# Assessment of listing and categorisation of animal diseases within the framework of the Animal Health Law (Regulation (EU) No 2016/429): West Nile fever

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# Assessment of listing and categorisation of animal diseases within the framework of the Animal Health Law (Regulation (EU) No 2016/429): West Nile fever

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## Abstract

West Nile fever (WNF) has been assessed according to the criteria of the Animal Health Law (AHL), in particular criteria of Article 7 on disease profile and impacts, Article 5 on the eligibility of WNF to be listed, Article 9 for the categorisation of WNF according to disease prevention and control rules as in Annex IV and Article 8 on the list of animal species related to WNF. The assessment has been performed following a methodology composed of information collection and compilation, expert judgement on each criterion at individual and, if no consensus was reached before, also at collective level. The output is composed of the categorical answer, and for the questions where no consensus was reached, the different supporting views are reported. Details on the methodology used for this assessment are explained in a separate opinion. According to the assessment performed, WNF can be considered eligible to be listed for Union intervention as laid down in Article 5(3) of the AHL. The disease would comply with the criteria as in Sections 2 and 5 of Annex IV of the AHL, for the application of the disease prevention and control rules referred to in points (b) and (e) of Article 9(1). The animal species to be listed for WNF according to Article 8(3) criteria are several orders of birds and mammals as susceptible species and several families of birds as reservoir. Different mosquito species can serve as vectors.



**Keywords:** West Nile fever, WNF, West Nile virus, WNV, Animal Health Law, listing, categorisation, impact

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# 1. Introduction

## 1.1. Background and Terms of Reference as provided by the requestor

The background and Terms of Reference (ToR) as provided by the European Commission for the present document are reported in Section 1.2 of the scientific opinion on the ad hoc methodology followed for the assessment of the disease to be listed and categorised according to the criteria of Article 5, Annex IV according to Article 9, and 8 within the Animal Health Law (AHL) framework (EFSA AHAW Panel, 2017a).

## 1.2. Interpretation of the Terms of Reference

The interpretation of the ToR is as in Section 1.2 of the scientific opinion on the ad hoc methodology followed for the assessment of the disease to be listed and categorised according to the criteria of Article 5, Annex IV according to Article 9, and 8 within the AHL framework (EFSA AHAW Panel, 2017a).

The present document reports the results of assessment on West Nile fever (WNF) according to the criteria of the AHL articles as follows:

- Article 7: West Nile fever profile and impacts
- Article 5: eligibility of West Nile fever to be listed
- Article 9: categorisation of West Nile fever according to disease prevention and control rules as in Annex IV
- Article 8: list of animal species related to West Nile fever.

# 2. Data and methodologies

The methodology applied in this opinion is described in detail in a dedicated document about the ad hoc method developed for assessing any animal disease for the listing and categorisation of diseases within the AHL framework (EFSA AHAW Panel, 2017a).

# 3. Assessment

## 3.1. Assessment according to Article 7 criteria

This section presents the assessment of WNF according to the Article 7 criteria of the AHL and related parameters (see Table 2 of the opinion on methodology (EFSA AHAW Panel, 2017a)), based on the information contained in the fact sheet as drafted by the selected disease scientist (see Section 2.1 of the scientific opinion on the ad hoc methodology) and amended by the AHAW Panel.

### 3.1.1. Article 7(a) Disease Profile

West Nile virus (WNV) belongs to the Flaviviridae family, genus *Flavivirus*, and is included in the serocomplex of Japanese Encephalitis virus together with Murray Valley encephalitis (MVE), St. Louis encephalitis (SLE), Kunjin (KUN), Usutu (USU), Koutango (KOU), Cacipacore (CPC), Alfuy (ALF) and Yaounde (YAO) viruses. Apart from Usutu virus, the other viruses in the serocomplex are not present in Europe. The virus was isolated for the first time in 1937 in Uganda, from the blood of a woman with febrile symptoms who came from the West Nile district (hence the name West Nile fever).

Different genetic lineages have been identified worldwide but the strains responsible for serious epidemics are attributable to Lineage 1 and, more recently, also to Lineage 2. Phylogenetic analyses revealed that all European WNV lineage 1 and 2 strains are derived from a limited number of independent introductions, most likely from Africa, followed by local spread and evolution. Other lineages have been identified but not associated so far with human or animal diseases.

WNV is transmitted by different genera and species of mosquitoes. The main vectors are some of the species of ornithophilic mosquitoes belonging to the genus *Culex*, which is always closely associated with the transmission of WNV during outbreaks. The mosquitoes cease their activity during the colder months, but it has been demonstrated that the virus is able to survive during this period in the infected mosquitoes, which overwinter indoors.

### 3.1.1.1. Article 7(a)(i) Animal species concerned by the disease

*Susceptible animal species*

Parameter 1 – Naturally susceptible wildlife species (or family/orders)

Several orders of birds can be naturally susceptible to WNV infections, i.e. Anseriformes, Apodiformes, Caprimulgiformes, Casuariiformes, Charadriiformes, Ciconiformes, Columbiformes, Coraciiformes, Cuculiformes, Falconiformes, Galliformes, Gaviformes, Gruiformes, Musophagiformes, Passeriformes, Pelecaniformes, Piciformes, Podicipediformes, Psittaciformes, Sphenisciformes, Strigiformes and Struthioniformes.

Also several orders of mammals can be naturally susceptible to WNV infections, i.e. Artiodactyla, Carnivora, Chiroptera, Perissodactyla, Primates, Proboscidea and Rodentia.

Two orders of reptiles can be naturally susceptible to WNV infections: Crocodylia and Squamata.

Details concerning the susceptible families and species of the above mentioned orders are listed in Table A.1 in Appendix A.

Parameter 2 – Naturally susceptible domestic species (or family/orders)

Several families of domestic animals can be naturally susceptible to WNV infections, i.e. Phasianidae, Anatidae, Bovidae, Canidae, Felidae, Leporidae and Equidae.

Details concerning the susceptible species of the above mentioned families are listed in Table A.2 in Appendix A.

Parameter 3 – Experimentally susceptible wildlife species (or family/orders)

Several wild birds of the orders Passeriformes, Falconiformes, Accipitriformes, Strigiformes, Galliformes, Pelecaniformes, Columbiformes, Gruiformes, Anseriformes, Charadriiformes, Psittaciformes and Piciformes were successfully infected (see Table A.3 in Appendix A for the outcomes of experimental infections of WNV performed in wild birds (adapted from Pérez-Ramírez et al. (2014) (Pérez-Ramírez et al., 2014)).

Parameter 4 – Experimentally susceptible domestic species (or family/orders)

Table A.4 in Appendix A lists the outcomes of experimental infections of WNV performed in domestic animal species. Infections have been successfully established in cats, dogs, horses, pigs, rabbits and sheep.

*Reservoir animal species*

Parameter 5 – Wild reservoir species (or family/orders)

Several bird species, particularly passerine species (jays, finches, sparrows, and crows) can be potential reservoirs of WNV. House finches (*Carpodacus mexicanus*) and house sparrows (*Passer domesticus*) experimentally inoculated showed persistent infection in spleen and kidney 28 weeks p.i. (post infection). The virus was still detected by real time reverse transcription polymerase chain reaction (RT-PCR) in the spleen of two house sparrows at 36 weeks p.i. However, viral isolation attempts were unsuccessful (Wheeler et al., 2012). In a previous work (Nemeth et al., 2009a), a higher number of organs were analysed in WNV-infected house sparrows, and viral RNA was detected in juvenile sparrows up to 65 days p.i in kidney and spleen, although infectious virus could be isolated at low titres only in one sparrow at 43 days p.i. Reisen et al. (2006) confirmed the persistent infection in five species of Passeriformes and in common ground-dove (*Columbina passerina*) detecting the virus in spleen and kidney, but also in lungs at > 6 weeks p.i. (Reisen et al., 2006).

Outside the United States of America (USA), clinical symptoms signs due to WNV infection have been reported in few cases and limited to scarce number of avian species in course of outbreaks: domestic geese (*Anser anser domesticus*) and white storks (*Ciconia ciconia*) during the WNV epidemic in Israel (Malkinson et al., 2002), goshawks (*Accipiter gentilis*) in Hungary (Bakonyi et al., 2006), Eurasian jays (*Garrulus glandarius*), little owl (*Athene noctua*), mallard (*Anas plathyrynchos*), and common buzzard (*Buteo buteo*) in Italy (Monaco et al., 2015). However, mass mortality of highly susceptible species (such as corvids or other species) is less frequently observed in the Old than in the New World although some species, as the jackdaws (*Corvus monedula*) could potentially function as sentinel (Lim et al., 2014). Surveillance activities carried out in Italy where WNV is endemic since 2008, pointed out the high susceptibility to the viral infection of three species of synantropic resident wild

birds, namely carrion crow (*Corvus corone*), magpie (*Pica pica*) and Eurasian jay (*Garrulus glandarius*) which justifies their use as sentinel in endemic areas (Italian Ministry of Health, 2016).

Some species of mammals including squirrels (*Sciurus* sp.), eastern chipmunks (*Tamias striatus*) and eastern cottontail rabbits (*Sylvilagus floridanus*) may be capable of transmitting WNV to mosquitoes, although their epidemiological role importance as reservoir hosts is still uncertain.

Among reptiles, clinical signs were mainly reported during outbreaks in alligators, although there is also a report on neurological signs associated with WNV infection in a crocodile monitor (*Varanus salvadori*) lizard. Some infections in garter snakes (*Thamnophis sirtalis*) experimentally inoculated with WNV were also fatal. Green iguanas (*Iguana iguana*) can be infected.

Amphibians including lake frogs (*Rana ridibunda*) and North American bullfrogs (*Rana catesbeiana*) can also be infected with WNV. Some alligators (e.g. American alligators, *Alligator mississippiensis*) and frogs (e.g. *Rana ridibunda* in Russia) may develop viraemia sufficient to infect mosquitoes. As with mammals, their epidemiological importance as reservoir hosts is still uncertain.

Based on preliminary research carried out in Italy and Spain, only a few bird species seem to play a major role in the transmission of infection to the mosquitoes (Hamer et al., 2009; Munoz et al., 2012; Roiz et al., 2012; Spedicato et al., 2016). Unfortunately, the reservoir competence for many European bird species is still unknown even though the persistence of WNV in infected birds has been assessed in some species through experimental trials. Table B.1 in Appendix B provides an overview of wild and domestic WNV reservoir/sentinel animal species.

Parameter 6 – Domestic reservoir species (or family/orders)

WNV has been associated with sporadic disease infection in small numbers of domestic animal species (see above in parameter 2 and Table A.2 in Appendix A); however, these species do not play a role in the further transmission of WNV to mosquitoes and are thus considered as dead-end hosts. See also Table B.1 in Appendix B which lists wild and domestic WNV reservoir/sentinel animal species.

#### 3.1.1.2. Article 7(a)(ii) The morbidity and mortality rates of the disease in animal populations

Parameter 1 – Prevalence/incidence

WNV has been found in all the continents from tropical to north temperate latitudes (Reisen, 2013). Table 1 lists the number of horses positive for WNV detections (either by immunoglobulin M enzyme-linked immunosorbent assay (IgM-ELISA) or PCR), reported to the Animal Diseases Notification System since between (ADNS) 2013 and 2016.

**Table 1:** Number of horses positive for WNV reported to the ADNS

| Country | Year | Number of positive horses |
|---|---|---|
| France | 2015 | 386 |
| Italy | 2013 | 341 |
| Italy | 2014 | 284 |
| Italy | 2015 | 339 |
| Italy | 2016 | 707 |
| Greece | 2013 | 575 |
| Greece | 2014 | 55 |
| Spain | 2013 | 330 |
| Spain | 2014 | 158 |
| Spain | 2015 | 309 |
| Spain | 2016 | 909 |
| Portugal | 2015 | 283 |
| Portugal | 2016 | 35 |
| Austria | 2016 | 17 |
| Hungary | 2013 | 12 |
| Hungary | 2014 | 91 |
| Hungary | 2015 | 92 |

| Country | Year | Number of positive horses |
|---|---|---|
| Hungary | 2016 | 945 |
| Bulgaria | 2015 | 2 |
| Bosnia and Herzegovina | 2013 | 0 |

Source: Animal Diseases Notification System (2013–2016).

Table C.1 in Appendix C summarises the prevalence of cases reported to the OIE in Europe, namely in Portugal, Spain, France, Croatia, Greece, Romania, Former Yugoslav Republic of Macedonia and Bulgaria. Also, the cases in Italy reported to the Italian authorities are summarised in Table C.1.

Parameter 2 – Case-morbidity rate (% clinically diseased animals out of infected ones)

WNF can cause disease in horses, several species of birds and rarely in other species such as camels, dogs, cats, sheep, squirrels and alligators (Go et al., 2014; Hubalek et al., 2014). In horses, the majority of the infections are asymptomatic, but some individuals (about 10%) can develop severe neurological illness (ataxia, weakness, recumbency and muscle fasciculation). Experimental infections have shown that the clinical picture of the disease can be quite divergent depending on the species. High susceptible species (e.g. corvids) can develop an hyperacute phase resulting in death without exhibiting symptoms, whereas other species (e.g. raptors, owls, Passeriformes) can develop only mild lesions with low mortality rates or chronic disease (Pérez-Ramírez et al., 2014). The case-morbidity rate in the outbreaks reported to the OIE and the Italian National Authorities are shown in Table C.1 in Appendix C.

Parameter 3 – Case-fatality rate

The case-fatality rate in the outbreaks in equids reported to the OIE and the Italian National Authorities are shown in Table C.1 in Appendix C.

LaDeau et al. (2007) demonstrated that the American crow population declined by up to 45% since WNV arrival in 1999 and only two of the seven species with documented impact recovered to pre-WNV levels by 2005 (LaDeau et al., 2007).

#### 3.1.1.3. Article 7(a)(iii) The zoonotic character of the disease

*Presence*

Parameter 1 – Report of zoonotic human cases (anywhere)

WNV zoonotic transmission is known to be present in Europe for many years: in the 1960s, the virus emerged in southern France in the Camargue. Yet, the first large outbreak in humans was reported from Bucharest, Romania in 1996–1997. Up to 2010, infection in humans and/or horses have been reported in the Czech Republic (1997), France (2000, 2003, 2004, 2006), Italy (1998, 2008, 2009), Hungary (2000–2009), Romania (1997–2001, 2003–2009), Spain (2004) and Portugal (2004). In 2010, a human outbreak was reported in northern Greece, and human cases were reported in Romania, Hungary, Italy, Spain and in Volgograd (Russian Federation). The number of human cases notified in Europe and in the Mediterranean Basin since 2010 is reported in Table 5.

#### 3.1.1.4. Article 7(a)(iv) The resistance to treatments, including antimicrobial resistance

Parameter 1 – Resistant strain to any treatment even at laboratory level

This is not applicable to WNV since there is no specific antiviral therapy.

#### 3.1.1.5. Article 7(a)(v) The persistence of the disease in an animal population or the environment

*Animal population*

Parameter 1 – Duration of infectious period in animals

Viral titres in blood equal or greater than $10^5$ $TCID_{50}$/mL have been considered able to infect competent mosquito species. In relation to viraemia duration, the following results of experimental infections in European bird species are reported:

**Table 2:** Duration of infection period in experimentally infected birds

| Species | Viraemia duration | Cloacal and oropharyngeal WNV shedding | Inoculum (WNV isolate, dose and inoculation route) | Challenge dose | Reference |
|---|---|---|---|---|---|
| Rock pigeons (*Columbia livia*) | 2 days (viraemia) | 15 dpi | 3 WNV Italian isolates (L1*) (IT/2009-IT/2011-IT/2012) 1 mL subcutaneously | $10^6$ $TCID_{50}$/mL | Spedicato et al. (2016) |
| Red-legged partridge (*Alectoris rufa*) | 4 days (viraemia) | 7 dpi | 1 WNV Morocco isolate (Mo/03) (L1) 1 WNV Spanish isolate (SP/07)(L1) 0.1 mL subcutaneously | $10^4$ PFU/bird | Sotelo et al. (2011b) |
| House sparrows (*Passer domesticus*) | 3 days (viraemia) | 12 dpi | 2 WNV Italian isolates (IT/2008 and IT/2009)(L1) 1 WNV Spanish isolate (SP/07)(L1) 1 WNV US isolate (NY99) (L1) 0.1 mL subcutaneously | $10^4$ PFU/bird | Del Amo et al. (2014) |
| Gyrfalcons (*Falco rusticolus*) | 4–6 days (viraemia) | 21 dpi | 1 WNV US isolate (NY99) (L1) 1WNV Austrian isolate (Aus/09)(L2*) 1 mL subcutaneously | Low dose: 500 $TCID_{50}$/mL Medium dose: $10^4$ $TCID_{50}$/mL High dose: $10^6$ $TCID_{50}$/mL | Ziegler et al. (2013) |

WNV: West Nile virus; $TCID_{50}$: tissue culture infective dose, median; PFU: plaque-forming unit.

Parameter 2 – Presence and duration of latent infection period

Evidence of persistent WNV infection has been demonstrated in experimentally infected monkeys (Pogodina et al., 1983) and hamsters (Tesh et al., 2005). WNV is also capable of long-term persistence in human patients, particularly in the presence of chronic clinical symptoms (Murray et al., 2010). The importance of these persistent infections, however, needs still to be elucidated, as virus titres are low and these hosts are considered to be dead-end hosts.

Parameter 3 – Presence and duration of the pathogen in healthy carriers

Refer to the data reported in Section 3.1.1.1 parameter 5.

*Environment*

Parameter 4 – Length of survival (dpi) of the agent and/or detection of DNA in selected matrices (soil, water, air) from the environment (scenarios: high and low T)

WNV is rapidly inactivated in the environment outside hosts. Low temperatures preserve infectivity, with stability being greatest below −60°C. It is inactivated by heat (50–60°C for at least 30 min), ultraviolet light, and gamma irradiation (Burke and Monath, 2001). The virus is also susceptible to disinfectants such as 3–8% formaldehyde, 2% glutaraldehyde, 2–3% hydrogen peroxide, 500–5,000 ppm available chlorine, alcohol, 1% iodine and phenol iodophors.

Data related to the persistence of the virus in the vectors are provided in Table 3.

**Table 3:** Detailed outcomes of systematic review on survival time of WNV in mosquitoes at different temperatures (data extracted from (Turell et al., 2002))

| Matrix | Target | Test | Temperatures | Maximum detection |
|---|---|---|---|---|
| Mosquito | Nucleic acid | RT-PCR | 4°, 20°, 70°C | 14 days |
| Mosquito | Virus | Culture | 4°, 20°, 70°C | 2 days |

RT-PCR: reverse transcription polymerase chain reaction.

### 3.1.1.6. Article 7(a)(vi) The routes and speed of transmission of the disease between animals, and, when relevant, between animals and humans

*Routes of transmission*

WNV is maintained in nature by a primary cycle of transmission mosquito–bird–mosquito (endemic cycle): adult ornithophilic mosquitoes (vectors) become infected by biting viraemic birds (amplifying hosts). A secondary cycle (epidemic cycle) is characterised by the involvement in the transmission cycle of accidental hosts such as horses and humans due to particular ecological conditions. In this case, arthropod vectors, called bridge vectors, are able to transmit the virus to hosts other than birds, such as horses and humans. Humans, equids and other mammals are considered to be dead-end accidental hosts. In these hosts, the virus does not reach a concentration in the bloodstream high enough to infect vectors, so the transmission cycle is not perpetuated. In Europe, the transmission cycle of WNV can be restricted to two main ecosystems: the rural (sylvatic) cycle, which occurs near wet/marshy areas between wild birds and ornithophilic mosquitoes, and the synanthropic/urban cycle, which arises between synanthropic or domestic birds and mosquitoes which can feed on the blood of birds and humans.

WNF vectors are mosquitos belonging to the *Culex*, *Aedes* and *Coquillettidia* genera (family *Culicidae*) (link to storymap VBD: https://efsa.maps.arcgis.com/apps/MapJournal/index.html?appid=512a03aa8df84d54a51bcb69d1b62735) (EFSA AHAW Panel, 2017b).

Parameter 1 – Types of routes of transmission from animal to animal (horizontal, vertical)

Results of experimental trials on WNV transmission routes in wild birds are summarised in Table 4 and Table A.3 in Appendix A.

Mosquito bites are the usual source of WNV for mammals, reptiles and amphibians; however, in some animals, there is also evidence for transmission by other routes. Carnivorous mammals and reptiles (e.g. cats and alligators) can be infected by eating contaminated tissues. Direct transmission during close contact has also been reported in alligators, possibly via faecal shedding of virus. Chipmunks, squirrels and raccoons can also shed WNV in faeces, oral secretions and/or urine. WNV has been found in the urine of experimentally infected hamsters, and in very small amounts in the oral and/or cloacal fluids of experimentally infected North American bullfrogs (*Rana catesbeiana*) and green iguanas (*Iguana iguana*). Transplacental transmission was reported in experimentally infected sheep and mice, as well as in a horse that was fatally infected with a Lineage 1 virus in Africa, and aborted in the final stage of the disease. The epidemiological significance (if any) of mammalian, reptilian and amphibian hosts in the maintenance or amplification of WNV remains to be established.

Parameter 2 – Types of routes of transmission between animals and humans (direct, indirect, including food-borne)

There is no evidence of natural direct transmission between vertebrates and humans. However, human infections from the exposure of conjunctival membranes (Fonseca et al., 2005) and/or percutaneous injury to the body fluids or tissues of WNV-infected birds (CDC, 2002) have been described.

**Table 4:** Experimental data on WNV transmission in wild birds

| Direct | Indirect* | Horizontal | Vertical | Species | Reference |
|---|---|---|---|---|---|
| C | Y | Y | NT | American crow (*Corvus brachyrhynchos*) | Komar et al. (2003) |
| C | Y | Y | NT | Blue jay (*Cyanocitta cristata*) | Komar et al. (2003) |
| C | Y | Y | NT | Black-billed magpie (*Pica hudsonia*) | Komar et al. (2003) |
| C | Y | Y | NT | Ring-billed gull (*Larus delawarensis*) | Komar et al. (2003) |
| C | Y | Y | N | Chicken (*Gallus gallus domesticus*)** | Langevin et al. (2001) |
| C | NT | Y | N | Domestic geese (*Anser anser domesticus*) | Swayne et al. (2001) |
| C | NT | | NT | Common goose (*Anser anser domesticus*) | Banet-Noach et al. (2003) |
| C | NT | Y | NT | Red-legged partridge (*Alectoris rufa*) | Sotelo et al. (2011b) |
| NT | Y | NT | NT | Canada goose (*Branta canadensis*) | Komar et al. (2003) |
| N | Y | N | NT | Mallard (*Anas platyrhynchos*) | Komar et al. (2003) |

| Direct | Indirect* | Horizontal | Vertical | Species | Reference |
|---|---|---|---|---|---|
| O | Y | Y | NT | American kestrel (*Falco sparverius*) | Komar et al. (2003) (C); Nemeth et al. (2006a) (O) |
| N | Y | N | NT | Northern bobwhite (*Colinus virginianus*) | Komar et al. (2003) |
| N | Y | N | NT | Japanese quail (*Coturnix japonicus*) | Komar et al. (2003) |
| NT | Y | NT | NT | Ring-necked pheasant (*Phasianus colchicus*) | Komar et al. (2003) |
| N | Y | N | NT | American coot (*Fulica americana*) | Komar et al. (2003) |
| NT | Y | NT | NT | Killdeer (*Charadrius vociferus*) | Komar et al. (2003) |
| N | Y | N | NT | Mourning dove (*Zenaida macroura*) | Komar et al. (2003) |
| N | Y | N | NT | Rock dove (*Columba livia*) | Komar et al. (2003) |
| N | Y | N | NT | Monk parakeet (*Myiopsitta monachus*) | Komar et al. (2003) |
| N | Y | N | NT | Budgerigar (*Melopsittacus undulatus*) | Komar et al. (2003) |
| O | Y | Y | NT | Great horned owl (*Bubo virginianus*) | Komar et al. (2003) (C); Nemeth et al. (2006a) (O) |
| NT | Y | NT | NT | Northern flicker (*Colaptes auratus*) | Komar et al. (2003) |
| N | Y | N | NT | Fish crow (*Corvus ossifragus*) | Komar et al. (2003) |
| N | Y | N | NT | American robin (*Turdus migratorius*) | Komar et al. (2003) |
| N | Y | N | NT | European starling (*Sturnus vulgaris*) | Komar et al. (2003) |
| NT | Y | NT | NT | Red-winged blackbird (*Agelaius phoeniceus*) | Komar et al. (2003) |
| N | Y | N | NT | Common grackle (*Quiscalus quiscula*) | Komar et al. (2003) |
| N | Y | N | NT | House finch (*Carpodacus mexicanus*) | Komar et al. (2003) |
| N | Y | N | NT | House sparrow (*Passer domesticus*) | Komar et al. (2003) |
| N | NT | N | NT | Red-tailed hawk (*Buteo jamaicensis*) | Nemeth et al. (2006a) |
| N | NT | N | NT | Song sparrow (*Melopiza melodia*) | Reisen and Fang (2007) |
| O | NT | Y | NT | Eastern screech owls (*Megascops asio*) | Nemeth et al. (2006b) |

C: Contact transmission; O: oral transmission; N: no evidence of direct transmission; NT: not tested.
*: Mosquitoes-exposed.
**: Only 1 animal in 16 in contact hens.

*Speed of transmission*

Transmission rate of WNV infection between vector (mosquito) and avian population has been expressed through the calculation of the basic reproduction number ($R_0$) by using different mathematical models. In the EU context, Calistri et al. (2016) developed a transitional mathematical model to calculate the $R_0$ values for the various part of the Italian territory from May to September, which resulted in a mean $R_0$ value for the whole Italy varying between 0.4 and 4.8, with values $> 1$ from the end of May to the beginning of September.

#### 3.1.1.7. Article 7(a)(vii) The absence or presence and distribution of the disease in the Union, and, where the disease is not present in the Union, the risk of its introduction into the Union

*Presence and distribution*

Parameter 2 – Type of epidemiological occurrence (sporadic, epidemic, endemic) at MS level

WNV introduction and circulation have been demonstrated on multiple occasions in southern Europe and in the Mediterranean basin since the 1960s when seropositive animals or virus isolates were discovered in France, Portugal and Cyprus (Filipe and Pinto, 1969; Joubert et al., 1970). Migratory birds have been associated with the introduction of viral strains from endemic areas (Calistri et al., 2010); however, the mechanism of virus persistence in animal hosts in Europe leading to endemicity of the infection is still unknown.

In Europe, WNV circulation was mainly detected in the Mediterranean and south-eastern regions, where notifications of human and horses cases of WNV infection have increased in the last 5–7 years, with the involvement of new areas, where the infection was not notified before, such as Bulgaria and Greece in 2010, Albania and Former Yugoslav Republic of Macedonia in 2011, and Croatia, Serbia and Kosovo in 2012. Accordingly, alarming outbreaks were reported in several European countries in 2010; 261 confirmed human cases, including 34 deaths, occurred in Greece, 57 cases and five deaths occurred in Romania, and 480 cases and six deaths occurred in Russia (Papa et al., 2010; Onishchenko et al., 2011; Sirbu et al., 2011).

Sporadic occurrence of the disease has been reported in France since 1962, when it first appeared in Camargue. In the same region, WNV was detected in 2000, 2004 and, after a ten-year period, in 2015 (Bahuon et al., 2016).

In Italy, WNV annual epidemics have been consistently registered since 2008 (Savini et al., 2008) caused by genetically divergent isolates and, to date, WNV is considered endemic in the north-eastern regions of the country, in Sardinia and in Sicily (Italian Ministry of Health, 2016).

The geographic distribution of West Nile cases in Europe and in Mediterranean Basin from 2008 to 2016 shown in Figure 1.

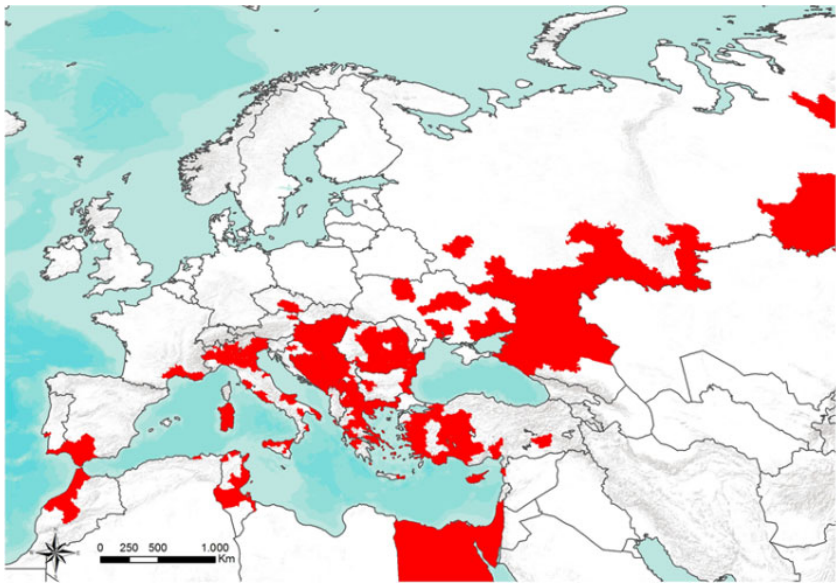


**Figure 1:** Geographic distribution of cases (confirmed and probable) of West Nile fever in Europe and in Mediterranean Basin (2008–2016) (source Arbozoonet: https://arbozoonet.izs.it/arbozoonet (ArboZoonet, online))

*Risk of introduction*

Data are not provided since the disease is already present in the Union. It should be noted, however, that a continuous introduction from Africa through migratory birds is suspected.

### 3.1.1.8. Article 7(a)(viii) The existence of diagnostic and disease control tools

*Diagnostic tools*

Parameter 1 – Existence of diagnostic tools

Details concerning the different types of diagnostic tools and their accuracy are listed in Table 7 in Section 3.1.4.1.

Viral nucleic acid and viral antigens can be demonstrated in tissues of infected animals by RT-PCR and immuno-histochemistry, respectively.

Antibodies can be detected in equine serum by IgM capture ELISA, haemagglutination inhibition (HI), IgG ELISA, plaque reduction neutralisation (PRN) or virus neutralisation (VN). In some serological assays, antibody cross-reactions with related flaviviruses, such as St. Louis encephalitis virus, Usutu virus, Japanese encephalitis virus or tick-borne encephalitis (TBE) virus may be encountered.

According to the OIE, the following tests are suitable methods for confirmation of clinical cases: Nested RT-PCR, real time RT-PCR and IgM capture ELISA. The PRN and serum neutralisation tests are

both suitable methods for detecting prevalence of infection, population freedom from infection and immune status in animals post-vaccination (Table 7).

Equine WNV-specific IgM antibodies are usually detectable from 7–10 days to 1–2 months post-infection. Most of horses with WNV encephalitis test positive in the IgM capture ELISA at the time that clinical signs are first observed. WNV neutralising antibodies are detectable in equine serum by 2 weeks post-infection and can persist for more than 1 year.

Several PCR methods are available as commercial kits. In view of the continued evolution and possible emergence of new WNV strains, it is important that the designs of PCR tests are constantly monitored and updated when necessary.

*Control tools*

Parameter 2 – Existence of control tools

In areas where the disease is endemic, horses may be protected from the clinical signs by vaccination (Table 7). In the infected areas, however, strategies aiming at reducing the circulation of the virus through the reduction of mosquito density (reduction/treatment of stagnant water, adulticidal and larvicidal targeted treatments) and of contacts between vectors and receptive hosts (application of repellent, mosquito netting, etc.) are the bases of any control policy for mosquito-borne diseases. Among biocidal products, the use of pyrethrin (6%) and piperonyl butoxide (60%) by aerial spray, indicated that the odds of infection after spraying were around six times higher in the untreated area than in treated areas, and that the treatments successfully disrupted the WNV transmission cycle (Carney et al., 2008).

### 3.1.2. Article 7(b) The impact of diseases

#### 3.1.2.1. Article 7(b)(i) The impact of the disease on agricultural and aquaculture production and other parts of the economy

*The level of presence of the disease in the Union*

Parameter 1 – Number of MSs where the disease is present

Since the beginning of the 2016 transmission season, the presence of WNV has been confirmed in MSs and neighbouring countries. As of 27 October 2016, 205 human cases of WNF have been reported in the EU and 261 cases in the neighbouring countries (Austria, Croatia, Cyprus, Egypt, Hungary, Italy, Israel, Portugal, Romania, Russian Federation, Serbia, Spain and Syrian Arab republic, Tunisia, Ukraine) (ECDC, 2016).

*The loss of production due to the disease*

Parameter 2 – Proportion of production losses (%) by epidemic/endemic situation

In European outbreaks, WNV has not been associated with any mortality in domestic birds but has been connected to a few cases in wild birds (see Section 3.1.1.1).

#### 3.1.2.2. Article 7(b)(ii) The impact of the disease on human health

*Transmissibility between animals and humans*

**Table 5:** Number of cases (confirmed and probable) of West Nile fever in Europe and in Mediterranean Basin (updated to 2 December 2016)

| Country | Year | Species | No. total cases[(a)] | No. confirmed cases[(b)] | Source |
|---|---|---|---|---|---|
| Albania | 2011 | Human | 2 | | ECDC (online) |
| Algeria | 2012 | Human | 1 | 1 | ECDC (online) |
| Austria | 2016 | Human | 2 | 2 | ECDC (online) |
| | 2015 | Human | 3 | 3 | |

| Country | Year | Species | No. total cases[a] | No. confirmed cases[b] | Source |
|---|---|---|---|---|---|
| | 2014 | Human | 1 | 1 | |
| Bosnia and Herzegovina | 2014 | Human | 13 | 0 | ECDC (online) |
| | 2013 | Human | 3 | 3 | |
| Bulgaria | 2016 | Human | 1 | 1 | ECDC (online) |
| | 2015 | Human | 2 | 0 | |
| Croatia | 2016 | Human | 1 | 0 | ECDC (online) |
| | 2013 | Human | 16 | 1 | ECDC (online) |
| | 2012 | Human | 5 | 3 | ECDC (online) |
| | 2013 | Horses | – | 12 | OIE (online) |
| Cyprus | 2016 | Human | 1 | 1 | ECDC (online) |
| Egypt | 2016 | Human | 1 | 1 | ECDC (online) |
| France | 2015 | Human | 1 | 1 | ECDC (online) |
| Former Yugoslav Republic of Macedonia | 2013 | Human | 1 | | ECDC (online) |
| | 2012 | Human | 6 | 1 | |
| Greece | 2014 | Human | 15 | 13 | HCDCP (online) |
| | 2014 | Horses | 4 | 4 | OIE (online) |
| | 2013 | Human | 86 | 58 | HCDCP (online) |
| | 2013 | Horses | – | 15 | OIE (online) |
| | 2012 | Human | 161 | 47 | HCDCP (online) |
| | 2012 | Horses | – | 15 | OIE (online) |
| | 2011 | Human | 101 | – | HCDCP (online) |
| | 2011 | Horses | 23 | – | OIE (online) |
| | 2010 | Human | 261 | – | HCDCP (online) |
| | 2010 | Horses | 30 | – | OIE (online) |
| Hungary | 2016 | Human | 39 | 16 | ECDC (online) |
| | 2015 | Human | 18 | 13 | ECDC (online) |
| | 2014 | Human | 11 | 3 | ECDC (online) |
| | 2013 | Human | 31 | 6 | ECDC (online) |
| | 2012 | Human | 12 | 7 | ECDC (online) |
| | 2011 | Human | 3 | – | ECDC (online) |
| | 2010 | Human | 3 | – | ECDC (online) |
| Israel | 2016 | Human | 80 | 47 | ECDC (online) |
| | 2015 | Human | 123 | 89 | |
| | 2014 | Human | 17 | 7 | |
| | 2013 | Human | 63 | 28 | |
| | 2012 | Human | 59 | 31 | |
| | 2011 | Human | 39 | – | |
| Italy | 2016 | Human | 71 | 71 | ISS (online) |
| | 2016 | Horses | 51 | 51 | IZSAM (online) |
| | 2015 | Human | 61 | 61 | ISS (online) |
| | 2015 | Horses | 30 | 30 | IZSAM (online) |
| | 2014 | Human | 24 | 24 | ISS (online) |
| | 2014 | Horses | 27 | 27 | IZSAM (online) |
| | 2013 | Human | 70 | 70 | ISS (online) |
| | 2013 | Horses | – | 50 | IZSAM (online) |
| | 2012 | Human | 50 | 39 | ISS (online) |
| | 2012 | Horses | – | 63 | IZSAM (online) |
| | 2011 | Human | – | 15 | ISS (online) |
| | 2011 | Horses | 197 | – | IZSAM (online) |

| Country | Year | Species | No. total cases[a] | No. confirmed cases[b] | Source |
|---|---|---|---|---|---|
| Kosovo | 2012 | Human | 4 | 0 | ECDC (online) |
| Former Yugoslav Republic of Macedonia | 2011 | Human | 4 | – | ECDC (online) |
| | 2011 | Horses | 10 | – | OIE (online) |
| Montenegro | 2013 | Human | 4 | – | ECDC (online) |
| | 2012 | Human | 1 | 1 | |
| Morocco | 2010 | Horses | 25 | – | OIE (online) |
| Palestine | 2014 | Human | 1 | 1 | ECDC (online) |
| | 2012 | Human | 2 | 1 | |
| Portugal | 2016 | Horses | 1 | 1 | OIE (online) |
| | 2015 | Human | 1 | 1 | ECDC (online) |
| | 2015 | Horses | 4 | 4 | OIE (online) |
| Romania | 2016 | Human | 93 | 80 | ECDC (online) |
| | 2015 | Human | 18 | 18 | ECDC (online) |
| | 2014 | Human | 23 | 22 | ECDC (online) |
| | 2013 | Human | 24 | 22 | ECDC (online) |
| | 2012 | Human | 14 | 13 | ECDC (online) |
| | 2011 | Human | 11 | – | ECDC (online) |
| | 2010 | Human | 52 | – | Sirbu et al. (2011) |
| | 2010 | Horses | 6 | – | OIE (online) |
| Russian Federation | 2016 | Human | 135 | 135 | ECDC (online) |
| | 2015 | Human | 39 | 39 | ECDC (online) |
| | 2014 | Human | 29 | – | ECDC (online) |
| | 2013 | Human | 177 | – | ECDC (online) |
| | 2012 | Human | 447 | – | ECDC (online) |
| | 2011 | Human | 153 | – | ECDC (online) |
| | 2010 | Human | 480 | – | Promed (online) |
| Serbia | 2016 | Human | 41 | 41 | ECDC (online) |
| | 2015 | Human | 28 | 28 | |
| | 2014 | Human | 76 | 56 | |
| | 2013 | Human | 302 | 200 | |
| | 2012 | Human | 70 | 41 | |
| Spain | 2016 | Human | 3 | 3 | Andalucía Ministry of Agriculture (online) |
| | 2016 | Horses | 70 | 70 | |
| | 2015 | Horses | 18 | 18 | |
| | 2013 | Horses | 40 | – | |
| | 2011 | Horses | 12 | – | |
| Syrian Arab Republic | 2016 | Human | 2 | 1 | ECDC (online) |
| Tunisia | 2016 | Human | 1 | 1 | ECDC (online) |
| | 2015 | Horses | 1 | 1 | OIE (online) |
| | 2013 | Human | 6 | 6 | ECDC (online) |
| | 2012 | Human | 63 | 33 | ECDC (online) |
| | 2011 | Human | 3 | – | ECDC (online) |
| Turkey | 2014 | Horses | 1 | 1 | OIE (online) |
| | 2011 | Human | 3 | – | ECDC (online) |
| | 2010 | Human | 7 | – | ECDC (online) |

| Country | Year | Species | No. total cases[(a)] | No. confirmed cases[(b)] | Source |
|---|---|---|---|---|---|
| Ukraine | 2016 | Human | 1 | 0 | ECDC (online) |
| | 2013 | Human | 1 | – | |
| | 2012 | Human | 12 | – | |
| | 2011 | Human | 8 | – | |

(a): For EU countries, probable and confirmed cases as per EU case definition (Commission Decision 2008/426/EC[1]).
(b): For EU countries, confirmed cases as per EU case definition (Commission Decision 2008/426/EC).

*Transmissibility between humans*

WNV is most commonly transmitted to humans by mosquitoes but additional routes of human-to-human transmission have also been documented as blood transfusions, organ transplants, exposure in a laboratory setting or the transmission from the mother to baby during pregnancy, delivery or breastfeeding. It is important to note that these methods of transmission represent a very small proportion of cases thus sufficient to evoke only a sporadic occurrence of the disease.

Humans are dead-end hosts since are not able to infect mosquitoes during the viraemic phase of the infection. Thus, the above-mentioned routes of direct transmission represent the main risk of infection dissemination among community. Laboratory acquired infections have also been reported (Campbell et al., 2002).

Parameter 3 – Human to human transmission is sufficient to sustain sporadic cases or community-level outbreak

WNV transmission through blood transfusion and organ transplantation is able to sustain community-level outbreak.

Parameter 4 – Sporadic, endemic, epidemic, or pandemic potential

Neuroinvasive human cases are usually sporadic, occurring mainly in immunocompromised persons or elderly.

*The severity of human forms of the disease*

Parameter 5 – Disability-adjusted life year (DALY)

Human infections are mostly asymptomatic. However, in some cases, they can exhibit a mild form of the disease (less than 1%) with encephalitis, meningoencephalitis or meningitis mainly among elderly or immunosupressed individuals (Go et al., 2014). As for most arthropod-borne diseases causing fever syndromes worldwide, the cumulative impact of WNV on global disease burden has not been fully assessed. Evaluations should include both the severe forms of the disease and the milder clinical manifestations which may result in neurological and ophthalmologic complications (Carson et al., 2006). WNV has been recognised able to induce a wide range of post-infection, long-term sequelae with the recovery of the affected patients within two years from the infection (Murray et al., 2008). However, a recent paper has emphasised that 40% of WNV-infected patients continued to experience symptoms related to their WNV infection up to 8 years later demonstrating the health and economic impact of a result of prolonged recovery, continued morbidity, and related disability (Murray et al., 2014).

*The availability of effective prevention or medical treatment in humans*

Parameter 6 – Availability of medical treatment and their effectiveness (therapeutic effect and any resistance)

There is no specific recommended treatment, other than supportive care, at present. Intensive care and mechanical ventilation may be required in some cases. Various therapies including interferon, antisense nucleotides and intravenous immunoglobulins (passive immunisation) are being tested in clinical trials. While a few case reports suggest that some of these treatments may be promising, larger studies are still lacking. Screening for new drugs that may inhibit WNV is underway.

[1] Commission Decision of 28 April 2008 amending Decision 2002/253/EC laying down case definitions for reporting communicable diseases to the Community network under Decision No 2119/98/EC of the European Parliament and of the Council (notified under document number C(2008) 1589) (Text with EEA relevance). OJ L 159, 18.6.2008, p. 46–90.

Parameter 7 – Availability of vaccines and their effectiveness (reduced morbidity)

There are no vaccines available for human use in EU.

#### 3.1.2.3. Article 7(b)(iii) The impact of the disease on animal welfare

Parameter 1 – Severity of clinical signs at case level and related level and duration of impairment

The incubation period for equine WNV encephalitis following mosquito transmission is estimated to be 3–15 days. A fleeting viraemia of low virus titre precedes clinical onset (Bunning et al., 2002). WNV encephalitis occurs in only a small per cent of infected horses; the majority of infected horses do not display clinical signs (Ostlund et al., 2000). The disease in horses is frequently characterised by mild to severe ataxia. Additionally, horses may exhibit weakness, muscle fasciculation and cranial nerve deficits (Cantile et al., 2000; Ostlund et al., 2000, 2001; Snook et al., 2001). Fever is an inconsistently recognised feature. Treatment is supportive and signs may resolve or progress to terminal recumbency. The mortality rate is approximately one out of three neurologically affected horses.

Many species of birds can become infected with WNV; the clinical outcome of infection is variable. Some species appear resistant while others suffer fatal neurologic disease. WNV infection associated with severe clinical signs have been described in several species of European wild birds (Bakonyi et al., 2006; Hofle et al., 2008; Jimenez-Clavero et al., 2008; Monaco et al., 2015).

#### 3.1.2.4. Article 7(b)(iv) The impact of the disease on biodiversity and the environment

*Biodiversity*

Parameter 1 – Endangered wild species affected: listed species as in CITES and/or IUCN list

CITES (online)

- *Phoenicopteridae* spp. (App. II)
- *Falco rusticolus* (App. I)
- *Aquila adalberti* (App. I)
- *Falconiformes* spp. (App. II)

Parameter 2 – Mortality in wild species

A number of outbreaks have been reported recently in Europe, Russia and parts of the Middle East. Since 2004, one introduced Lineage 2 virus in Central Europe has affected significant numbers of wild and captive raptors (Erdélyi et al., 2007). Therefore, the potential for WNV to cause illness or deaths in other European birds should be re-examined. Some virus lineages seem to have become endemic and are spreading (CFSPH, 2013). Species known to be susceptible to this isolate include sparrow hawks (*Accipiter nisus*), goshawks (*Accipiter gentilis*) and gyrfalcons (*Falco rusticolus*). The same virus was isolated from a dead collared dove (*Streptopelia decaocto*) in Italy, during an outbreak characterised by observed of mortality in collared doves and other species, including blackbirds. Different lineages of the WNV have also been found occasionally in other dead birds including European robins (*Erithacus rubecula*), raven (*Corvus corax*), common magpies (*Pica pica*), Eurasian jay (*Garrulus glandarius*), house sparrows (*Passer domesticus*), black redstart (*Phoenicurus ochruros*), sedge warbler (*Acrocephalus schoenobaenus*) and Savi's warbler (*Locustella luscinioides*).

LaDeau et al. (2007) demonstrated a high impact on the abundance of seven species of North American wild birds after the emergence of WNV in 1999. Host susceptibility, spatio-temporal heterogeneity in pathogen transmission and other environmental impacts on populations were accounted for using Bayesian modelling techniques. These seven species included two members of the family Corvidae (American crow and blue jay), two from Turdidae (American robin and eastern bluebird), two from Paridae (chickadees and tufted titmouse) and one from Troglodytidae (house wren). Also, George et al. (2015) demonstrated significant negative effects on survival of 47–49% bird species in North America, using an extensive capture-recapture technique study of nearly two decades, combined with recently developed models of WNV risk (George et al., 2015). The authors suggested that WNV in the US has a significant persistent effect on wild bird populations long after initial concerns had stopped.

### 3.1.3. Article 7(c) Its potential to generate a crisis situation and its potential use in bioterrorism

Parameter 1 – Listed in OIE/CFSPH classification of pathogens

WNV is listed in the CDC list of potential bioterrorism agents.

Parameter 2 – Listed in the Encyclopaedia of Bioterrorism Defence of Australia Group

WNV is not listed in the Encyclopaedia of Bioterrorism Defence of Australia Group.

Parameter 3 – Included in any other list of potential bio- agro-terrorism agents

WNV is not reported in any other list of potential bio-agro-terrorism agents.

### 3.1.4. Article 7(d) The feasibility, availability and effectiveness of the following disease prevention and control measures

#### 3.1.4.1. Article 7(d)(i) Diagnostic tools and capacities

*Availability*

Parameter 1 – Officially/internationally recognised diagnostic tool, OIE certified

**Table 6:** Test methods available for the diagnosis of WNV and their purpose (Source: (OIE, 2013))

| Method | Purpose | | | | |
|---|---|---|---|---|---|
| | Population freedom from infection | Individual animal freedom from infection | Confirmation of clinical signs | Prevalence of infection | Immune status in individual animals or populations post-vaccination |
| **Agent identification** | | | | | |
| Nested RT-PCR | – | ** | ** | – | – |
| Real-time RT-PCR | – | ** | ** | – | – |
| Isolation in tissue culture | – | ** | ** | – | – |
| **Detection of immune response** | | | | | |
| IGM capture ELISA | – | – | ** | – | |
| Plague reduction neutralisation | ** | – | * | ** | ** |
| Serum neutralisation | ** | – | * | ** | ** |
| Immunohistochemistry | – | – | * | – | |

*** = recommended method; ** = suitable method; * = may be used in some situations, but cost reliability, or other factors severity limits its application; – = not appropriate for this purpose.
Although not all of the rests listed as category *** have undergone formal validation, their routine nature and the fact that they have been used widely without dubious results, makes them acceptable.
RT-PCR: reverse transcriptase polymerase chain reaction; IgM: immunoglobulin M; ELISA: enzyme linked immunosorbent assay.

*Effectiveness*

Parameter 2 – Se and Sp of diagnostic test

Diagnostic tests, their type, accuracy and basis for WNF diagnosis are listed in Table 7.

**Table 7:** Diagnostic tests for WNV

| Test | Target | Se | Sp | Matrix | Reference | Notes |
|---|---|---|---|---|---|---|
| NS1-antigen protein microarray | Antibodies | 95% | 100% | Serum | Cleton et al. (in press) | Differential diagnosis of flavivirus infections in horses |
| Real-time RT-PCR | Antigen | From 1.5 to 15 copies per reaction | 100% | Viral strains, human samples (cerebrospinal fluid, biopsies, serum and plasma) and mosquito pools | Vázquez et al. (2016) | Specificity evaluated using viral RNA from a panel of different flaviviruses and other encephalitic viruses belonging to several viral families |

| Test | Target | Se | Sp | Matrix | Reference | Notes |
|---|---|---|---|---|---|---|
| Real-time RT-PCR | Antigen | 80 genome copies | 100% | Viral strains Lineages 1 and 2 | Faggioni et al. (2014) | Specificity evaluated using TBE, Usutu, Dengue 1, Dengue 4, YF, JEV |
| SYBR Green I-based real-time RT-PCR | Antigen | 20 copies | 100% | Human serum/plasma | Kumar et al. (2014) | Specificity evaluated using DEN-1–4, JEV, YFV, SLEV |
| Antigen capture ELISA | Antigen | 90% | 98% | Human serum | Saxena et al. (2013) | Detection of NS1 antigen |
| Real-time RT-PCR | Antigen | 10 copies | 100% | Viral strains | Barros et al. (2013) | Detection and differentiation between WNV and JEV; specificity evaluated using DEN-1–4, JEV, YFV, ZIKAV, Ntaya, TBEV, USUV, Toscana, CHIKV |
| Real-time RT-PCR | Antigen | 1.26 $TCID_{50}$/ml for WNV-L1, 6.3 $TCID_{50}$/ml for WNV-L2 | 100% | Tissue, feathers, oropharyngeal and cloacal swabs and blood from wild birds, samples from mice infected experimentally | Del Amo et al. (2013) | Detection and differentiation between WNV and USUV; specificity evaluated using SLEV, MVEV, JEV, BAGV, DEN-1, TBEV, VEEV, VSV, AIV, EIV, NDV, AHS4 |
| Competitive ELISA | Antibodies | 100% | Wild birds: 79.5% compared to VNT | Sera from mammals and wild birds | Sotelo et al. (2011a) | |
| | | | Horses: 96.5% compared to VNT | | | |
| | | | South african mammals: 79.5% compared to HAI | | | |
| | | | Giraffes: 67% compared to HAI | | | |
| IgM capture ELISA | Antibodies | 91.7% | 99.2% | Horse sera | Long et al. (2006) | |
| Real-time RT-PCR | Antigen | 2–4 genome copies of WNV | 100% | Viral strains | Eiden et al. (2010) | In OIE manual. For simultaneous detection and differentiation of WNV Lineage 1 and Lineage 2. Specificity evaluated using TBEV, YFV, JEV |
| Nested RT-PCR | Antigen | 10–8.0/100 µL | ND | Equine brain, blood, and cerebrospinal fluid; avian brain tissues | Johnson et al. (2001) | In OIE manual |

| Test | Target | Se | Sp | Matrix | Reference | Notes |
|---|---|---|---|---|---|---|
| Real-time RT-PCR | Antigen | 0.1 PFU | 100% | Human serum, CSF, brain tissue, mosquito pools, and avian tissues | Lanciotti et al. (2000) | In OIE manual. Specificity evaluated using DEN-2, JEV, YFV, SLEV, Lacrosse virus, Powassan virus, MVE, WEEV, EEEV |

RT-PCR: reverse transcriptase polymerase chain reaction; IgM: immunoglobulin M; ELISA: enzyme linked immunosorbent assay; PFU: plaque-forming unit.

*Feasibility*

Parameter 3 – Type of sample matrix to be tested (blood, tissue, etc.)

See Table 7.

### 3.1.4.2. Article 7(d)(ii) Vaccination

WNV vaccines approved by EMA are listed in Table 8.

**Table 8:** Vaccines for horses authorised for commercialisation in the EU by the European Medicines Agency (updated in October 2016) and their efficacy as emerged from a systematic review (updated to January 2016)

| Commercial name of vaccine | Type of vaccine | Way of administration | Doses | Species for which authorised | Countries in which authorised | Manufacturer | Efficacy | Field protection | Yearly availability/ production capacity | Ref. |
|---|---|---|---|---|---|---|---|---|---|---|
| **Proteq West Nile** | West Nile recombinant canarypox virus, vCP2017 virus | IM | | Horses | All EU | Merial | NA | NA | NA | |
| **Equilis West Nile** | Inactivated chimaeric flavivirus strain YF-WN | IM | | Horses | All EU | Intervet International BV | NA | NA | NA | |
| **Equip WNV (previously Duvaxyn WNV)** | Inactivated West Nile virus, strain VM-2 | IM | 2 doses (21 days apart) | Horses | All EU | Zoetis Belgium SA | Viruses could be isolated from 8 out of 10 non-vaccinated animals up to 14 days after challenge, but only 1 vaccinated animals. Sixty per cent of the controls had to be euthanised after challenge compared to none of the vaccinates. From 10 non-vaccinated animals, all presented, up to 21 days after challenge, pyrexia, head tremors or muscle fasciculations, and anxiety, and 9 showed mild paresis. In controls, these numbers were 2, 2, 6 and 2, respectively | Experimental trial | NA | Bowen et al. (2014) |

NA: data not available; IM: intramuscular.

#### 3.1.4.3. Article 7(d)(iii) Medical treatments

There is no specific recommended treatment, other than supportive care, at present.

#### 3.1.4.4. Article 7(d)(iv) Biosecurity measures

The biosecurity measures aiming at reducing the WNV spread are focused on controlling the vectors primarily responsible for the viral transmission. Farm-to-farm movement of infected horses is not effective to spread the disease since they are neither able to transmit the virus to biting mosquitoes nor, directly, to vertebrates including humans.

To minimise the possibilities of contact between the vectors and receptive hosts, it is advisable to use mosquito nets to avoid the vector entrance in the stables as well as the use of repellents on the animals. Data related to the efficacy of these substances has been detailed in Section 3.1.5.4 parameter 1.

To prevent any inter-human spread, the screening of blood and organs for transplantation in areas with WNV circulation is a common measure.

#### 3.1.4.5. Article 7(d)(v) Restrictions on the movement of animals and products

No specific measures are mentioned in the EU legislation for WNV outbreak control.

#### 3.1.4.6. Article 7(d)(vi) Killing of animals

No specific measures are mentioned in the EU legislation for WNV outbreak control.

#### 3.1.4.7. Article 7(d)(vii) Disposal of carcasses and other relevant animal by-products

No specific measures are mentioned in the EU legislation for WNV outbreak control.

### 3.1.5. Article 7(e) The impact of disease prevention and control measures

#### 3.1.5.1. Article 7(e)(i) The direct and indirect costs for the affected sectors and the economy as a whole

The major impact of WNV on animal health in the EU ecosystem is limited to the development of clinical signs in horses and, to date, there are no reports of clinical illness in domestic bird species. Thus, the major costs of WNV control in animals, namely horses, should include:

a) the cost of vaccination: primary vaccination consists of two doses, the second dose being administered 3–6 weeks later, depending on the vaccine used;
b) the cost to prevent mosquitoes bites: keeping horses indoor is not be very effective against *Culex pipiens* if additional measures such as mosquito nets or fans are not installed, as these mosquitoes are also active indoors. The use of insecticides or repellents is also able to reduce the possibilities for contact between the vectors and receptive hosts. Control of vectors can be recommended to individuals and to public health authorities in case of a severe epidemic, but the associated costs are difficult to estimate since emergency aerial spraying, even if proven to be effective in reducing mosquito populations and the number of human cases of WNV infection in the US (Barber et al., 2010), would not be the first option of vector control in MSs, given the substantial environmental risks and not easily accepted by the population (Humblet et al., 2016).
c) the costs of active surveillance activities, which may vary considerably between MSs. Usually animal surveillance encompasses domestic solipeds (horses and donkeys), birds and other animal species (e.g. cattle and farmed deer), as well as entomological surveillance activities. The main objective of the surveillance in humans during the transmission period is to ensure an immediate response in the implementation of the blood safety measures and the prevention of human cases, and, on an annual basis, to improve and adapt the surveillance and strengthen the preparedness.
d) WNV RNA screening of all blood donors in areas where the WNV circulation is in place. As an example in Italy during the 2015 epidemics, a total of 316,614 WNV NAT screening tests were conducted in blood donors in the affected provinces and 13 asymptomatic infected donors, were identified. No donor or organ transplant recipients were positive for WNV among the 168 tested.

#### 3.1.5.2. Article 7(e)(ii) The societal acceptance of disease prevention and control measures

The control of the mosquito population through the intensive use of biocidal products, e.g. by aerial spray is not easily accepted by the population (Humblet et al., 2016).

#### 3.1.5.3. Article 7(e)(iii) The welfare of affected subpopulations of kept and wild animals

Parameter 1 – Welfare impact of control measures on domestic animals

Since no specific measures are mentioned in the EU legislation for the WNV outbreak control, there is no impact on the welfare of domestic animals of official control measures.

Parameter 2 – Wildlife depopulation as control measure

Wild bird depopulation is not a control measure applied in course of WNV outbreak and its efficacy, as emerged from epidemiological models, not ascertained since the potential reduction of bird densities could enhance WNV transmission (Wonham et al., 2004).

#### 3.1.5.4. Article 7(e)(iv) The environment and biodiversity

*Environment*

Parameter 1 – Use and potential residuals of biocides or medical drugs in environmental compartments (soil, water, feed, manure)

In WNV-infected areas strategies must be implemented to reduce the circulation of the virus through measures that modify the density of the vectors (reduction of stagnant water, performance of adulticidal and larvicidal treatments) and to reduce the possibilities of contact between the vectors and receptive hosts (application of repellent, mosquito netting, etc.). Among biocidal products, the use of pyrethrin (6%) and piperonyl butoxide (60%) by aerial spray indicated that the odds of infection after spraying were around 6 times higher in the untreated area than in treated areas, and that the treatments successfully disrupted the WNV transmission cycle (Carney et al., 2008). Since *Cx. pipiens* is considered to be the main vector of WNV in Europe a list of biocidal products targeting mosquito control are reported in Table 9.

**Table 9:** Biocidal products targeting mosquito control (genus *Culex*), for which reports were found in a systematic review of available treatments against the vectors of vector-borne infections (papers published up to January 2016)

| Active substance | Reference | Intended use (route investigated in the study) | Study findings |
|---|---|---|---|
| **Studies not targeting any particular host** | | | |
| **Deltamethrin** | Marcombe et al. (2011) | **Fogging**<br>Vehicle-mounted thermal foggers (1 g/ha) | Efficacy was assessed by monitoring mortality rates of naturally resistant and laboratory susceptible mosquitoes placed in sentinel cages. Results showed high mortality rates of susceptible sentinel mosquitoes (64%) while resistant mosquitoes exhibited very low mortality (10%) |
| **Studies focused on vector control in housing/environment** | | | |
| **Deltamethrin** | Akogbéto et al. (2010) | **Indoor spraying** | Deterrence rate[(a)]: *Anopheles gambiae* (31.25%, 24.75%, 30 and 60 dpt; *Culex* sp. and *Mansonia* sp. 30 dpt 46.15%) |
| | | Huts were treated with insecticides. The absorption of the walls was 112 mL of insecticide per $m^2$ and that of the ceiling (polyethylene), the entry slits, and the door (painted metal) was in total 53.13 mL/$m^2$ | Exophily rate[(b)]: *Anopheles gambiae* (45.4%, 26.3%, 30 and 60 dpt; *Culex* sp. and *Mansonia* sp. 30 dpt 33.3%) |
| | | | Blood-feeding rate[(c)]: *Anopheles gambiae* (18.2%, 23.7%, 30 and 60 dpt; *Culex* sp. and *Mansonia* sp. 30 dpt, 14.3%) |
| | | | Immediate mortality[(d)]: *Anopheles gambiae* (32.7%, 15.8%, 30 and 60 dpt; *Culex* sp. and *Mansonia* sp. 30 dpt, 8.5%) |

| Active substance | Reference | Intended use (route investigated in the study) | Study findings |
|---|---|---|---|
| | | | Overall mortality[(e)]: *Anopheles gambiae* (72.7%, 31.6%, 30 and 60 dpt; *Culex* sp. and *Mansonia* sp. 30 dpt, 21%) |
| **Deltamethrin** | Badolo et al. (2014) | | Mortality of mosquitoes was 90.5 (86–94)% in unwashed nets (3 min exposure, 24-h mortality), and remained above 90% after 5 washes. Average mortality after 10, 15 and 20 washes were 81 (75–86)%, 68.7 (63–75)% and 66.3 (60–72)%, respectively |
| **Deltamethrin** | Dabiré et al. (2006) | **Treated mosquito nets**<br>Concentration of 55 mg/m$^2$ | Mosquito entrance rate was 10-fold higher in control houses than in houses with long lasting impregnated nets (LLINs) and there was no difference between the two tested net types. Among mosquitoes found in the houses, 36% were dead in LLIN houses compared to 0% in control houses. Blood feeding rate was 80% in control houses compared to 43% in LLIN houses. The type of net did not significantly impact any of these parameters |
| **Deltamethrin** | Darriet et al. (2000) | **Treated mosquito nets**<br>Concentration of 25 mg/m$^2$ | The 24-h mortality was 56% for *Anopheles gambiae* females, and 45% for *Culex* spp. females (compared to 4 and 6% in controls) |
| **Deltamethrin** | Moosa-Kazemi et al. (2007) | **Treated mosquito nets**<br>Concentrations of 25 mg/m$^2$ | Recorded 24-h-mortality was 100% even after 9 months |
| **Deltamethrin** | Muller et al. (2002) | **Treated mosquito nets**<br>Concentrations from 55 mg/m$^2$ (unwashed) to 1.6 mg/m$^2$ (18 months old and washed 3 times) | Mortality of mosquitoes was 97% in washed nets, and reduced to 84%, 54% and 7% after 6, 12 and 18 months (with respective average of times washed of 1.1, 1.9 and 3) |
| **Deltamethrin** | Van Roey et al. (2014) | **Treated mosquito nets**<br>Concentrations of 55 and 68 mg/m$^2$ | A positive control (commercial product PermaNet® 2.0, 55 mg a.i./m$^2$) was able to kill over 90% of mosquitoes (3 min exposure, 24-h-mortality) for up to 30 months, while the observed mortality with the experimental product (Netprotect®, 68 mg a.i./m$^2$) was 85.7% after 12 months, and remained below 90% |
| **Diflubenzuron** | Cetin et al. (2006) | **Septic tank water treatment**<br>0.01, 0.02, and 0.03 mg (AI)/L, using a 25% wettable powder or a 4% granular formulation in wastewater tank | Recorded adult inhibition for *Culex pipiens* was always 100% in the first 2 weeks, for all concentrations tested, and remained at 100% for up to 4 weeks with 30 g/L, and 2 weeks with 10 g/L |
| **Lambda-cyhalothrin** | Okumu et al. (2012) | **Indoor spraying**<br>0.03 g/m$^2$ sprayed on mud walls | Mortality (24-h mortality of *Anopheles arabiensis*) was 90% after 30 days but reduced to 35% after 60 days |
| **Lambda-cyhalothrin** | Trout et al. (2007) | **Outdoors Spraying** | The reduction in *Aedes albopictus* in sites was of 89.5% compared to controls, and in laboratory bioassays exposing mosquitoes to treated |

| Active substance | Reference | Intended use (route investigated in the study) | Study findings |
|---|---|---|---|
| | | Mist (concentration of 62.52 mL/L) directly applied to vegetation in the backyard of houses, and other resting sites | leaves, mortality varies from 80% after 2 weeks, to 35% after 8 weeks. In contrast, *Culex* spp. were not reduced |
| **Permethrin** | Rozendaal et al. (1989) | **Treated mosquito nets** | Cotton cloth impregnated with permethrin at a rate of 0.5 g/m$^2$ killed all *Anopheles darlingi* females exposed for 2 min, but after the material had been washed twice in soapy water the bioassay mortality fell to only 21.4%. Bioassays with *Culex quinquefasciatus* females showed that sprayed nets were less effective than nets impregnated by soaking (at equivalent dosages of 0.16–1.34 g/m$^2$) |
| | | Concentrations of 125–1,000 mg/m$^2$ | |
| **Permethrin** | Soleimani-Ahmadi et al. (2012) | **Treated mosquito nets** | Mortality of mosquitoes was 100% in the first 90 days, 92.4% (88–97) after 5 months, and reduced to 81.6% (75–88) after 9 months, and 72.3% (65–79) after 12 months |
| | | The nets were blended with 1,000 mg a.i/m$^2$ (2%, w/w), and final concentrations varied from 814 to 937 mg/m$^2$ | |
| **Studies focused on humans as the host species (personal protection)** | | | |
| DEET | Soonwera and Phasornkusolsill (2015) | External use – topic/ spray | DEET was used as control when evaluating other (non-ECHA approved) substances. The formulation gave protection for up to 182 min, and 98.5% protection from bites of *Aedes aegypti* and *Culex quinquefasciatus* |
| | | DEET 20% (w/w), 0.1 mL applied on a 3 × 10 cm area on the ventral portion of the forearm | |
| DEET | Gupta et al. (1987) | **Treated clothes and topic applications of repellent**, in different concentrations and combinations | The field trials were arranged in a four-way factorial design which compared fabric types, permethrin treatment and repellent treatments over a 14-h test period. The repellent formulations and the permethrin-treated clothing used as one system provided better protection (81% mortality) than the repellent formulations or permethrin-treated clothing used separately |
| DEET + permethrin | Mani et al. (1991) | **External use – soap** | Percentage repellency (reduction in biting rates) was 96% for *Culex vishnui*, 89.6% for *Culex tritaeniorhynchus* and 94.8% for *Culex pseudouishnui* |
| | | Containing 20% DEET and 5% permethrin | |
| Metofluthrin | Dame et al. (2014) | 'Clip-on' **spatial repellent device** | Efficacy in reduction of *Anopheles quadrimaculatus*, in 2 study years, compared to control, were 16% and 8%) |
| | | 31.20% | 19% and 8% for *Psorophora columbiae* and 69% for *Culex erraticus*. Total mosquito reduction was 13% |
| Metofluthrin | Revay et al. (2013) | **External use** | Biting on the arms of volunteers was reduced by 96.28% for *Ae. albopictus*, and by 94.94% for *Cx. pipiens* |
| | | 'Clip-on' metofluthrin (31.2%) | |

(a): Percentage of reduction in the number of mosquitoes caught in treated hut relative to the number caught in the control hut.
(b): Percentage of mosquitoes that have escaped the hut and have taken refuge in the veranda trap divided by the total number of mosquitoes collected in the hut.
(c): Percentage of blood fed mosquitoes collected divided by the total of mosquitoes collected in veranda and hut.
(d): Percentage of dead mosquitoes collected in the morning compared to total mosquitoes collected in the hut.
(e): Immediate mortality plus delayed mortality recorded after 24 h.

*Biodiversity*

Parameter 2 – Mortality in wild species

The main risk may be represented by the environmental residual of biocides which may interfere with ecology of wild species.

## 3.2. Assessment according to Article 5 criteria

This section presents the results of the expert judgement on the criteria of Article 5 of the AHL about WNF (Table 10). The expert judgement was based on Individual and Collective Behavioural Aggregation (ICBA) approach described in detail in the opinion on the methodology (EFSA AHAW Panel, 2017a). Experts have been provided with information of the disease fact-sheet mapped into Article 5 criteria (see supporting information, Annex A), based on that the experts indicate their Y/N or 'na' judgement on each criterion of Article 5, and the reasoning supporting their judgement.

The minimum number of judges in the judgement was 12. The expert judgement was conducted as described in the methodological opinion (EFSA AHAW Panel, 2017a). For details on the interpretation of the questions, see Appendix B of the methodological opinion (EFSA AHAW Panel, 2017a).

**Table 10:** Outcome of the expert judgement on the Article 5 criteria for West Nile fever

| **Criteria to be met by the disease:** According to AHL, a disease shall be included in the list referred to in point (b) of paragraph 1 of Article 5 if it has been assessed in accordance with Article 7 and meets all of the following criteria | | **Final outcome** |
|---|---|---|
| A(i) | The disease is transmissible | Y |
| A(ii) | Animal species are either susceptible to the disease or vectors and reservoirs thereof exist in the Union | Y |
| A(iii) | The disease causes negative effects on animal health or poses a risk to public health due to its zoonotic character | Y |
| A(iv) | Diagnostic tools are available for the disease | Y |
| A(v) | Risk-mitigating measures and, where relevant, surveillance of the disease are effective and proportionate to the risks posed by the disease in the Union | Y |
| **At least one criterion to be met by the disease:** In addition to the criteria set out above at points A(i)-A(v), the disease needs to fulfil at least one of the following criteria | | |
| B(i) | The disease causes or could cause significant negative effects in the Union on animal health, or poses or could pose a significant risk to public health due to its zoonotic character | Y |
| B(ii) | The disease agent has developed resistance to treatments and poses a significant danger to public and/or animal health in the Union | na |
| B(iii) | The disease causes or could cause a significant negative economic impact affecting agriculture or aquaculture production in the Union | N |
| B(iv) | The disease has the potential to generate a crisis or the disease agent could be used for the purpose of bioterrorism | NC |
| B(v) | The disease has or could have a significant negative impact on the environment, including biodiversity, of the Union | NC |

Colour code: green = consensus (Yes/No); yellow = no consensus (NC); red = not applicable (na), i.e. insufficient evidence or not relevant to judge.

### 3.2.1. Non-consensus questions

This section displays the assessment related to each criterion of Article 5 where no consensus was achieved in form of tables (Tables 11 and 12). The proportion of Y, N or na answers are reported, followed by the list of different supporting views for each answer.

**Table 11:** Outcome of the expert judgement related to criterion 5 B(iv)

| Question | | Final outcome | Response | | |
|---|---|---|---|---|---|
| | | | Y (%) | N (%) | na (%) |
| B(iv) | The disease has the potential to generate a crisis or the disease agent could be used for the purpose of bioterrorism | NC | 83 | 17 | 0 |

NC: non-consensus; number of judges: 12.

Reasoning supporting the judgement

Supporting Yes:

- It is listed in OIE/CFSPH, there is public concern on the disease and the potential to create a crisis.
- There have been examples of public health crisis in Romania in the 1990s, in Greece in 2010, in Hungary and Russia following outbreaks in humans.
- US army indicates that virulent genes could be modified to increase pathogenicity for humans and used as a weapon (since transmitted by mosquitoes).
- There were crisis in naïve areas, but not in endemic areas like France.

Supporting No:

- Some MSs do not have any WNV monitoring system in place, while others have been operating systems for several years, e.g. Italy and Greece. The main objective of the surveillance in humans during the transmission period is to ensure an immediate response in the implementation of the blood safety measures and the prevention of human cases, and, on an annual basis, to improve and adapt the surveillance and strengthen the preparedness. In Italy, for example, though the repeated and constant WNV circulation, surveillance on blood samples has been put in place and this also has not generated a crisis. Moreover, WNV circulation in France have not generated crisis.
- The situation in the US is not related with bioterrorism.
- Every virus could be genetically modified and become a threat, a worst-case scenario exists for every disease and should not be considered here.

**Table 12:** Outcome of the expert judgement related to criterion 5 B(v)

| Question | | Final outcome | Response | | |
|---|---|---|---|---|---|
| | | | Y (%) | N (%) | na (%) |
| B(v) | The disease has or could have a significant negative impact on the environment, including biodiversity, of the Union | NC | 82 | 18 | 0 |

NC: non-consensus; number of judges: 11.

Reasoning supporting the judgement

Supporting Yes:

- The North American experience shows that there is the potential for significant impact on the biodiversity.

Supporting No:

- There is no report of an impact of WNF at population level on endangered species in EU.

### 3.2.2. Outcome of the assessment of West Nile fever according to criteria of Article 5(3) of the AHL on its eligibility to be listed

As from the legal text of the AHL, a disease is considered eligible to be listed as laid down in Article 5 if it fulfils all criteria of the first set from A(i) to A(v) and at least one of the second set of criteria from B(i) to B(v). According to the assessment methodology (EFSA AHAW Panel, 2017a), a criterion is considered fulfilled when the outcome is 'Yes'. According to the results shown in Table 10, WNF complies with all criteria of the first set and with one criterion of the second set, therefore it is considered eligible to be listed as laid down in Article 5 of the AHL.

## 3.3. Assessment according to Article 9 criteria

This section presents the results of the expert judgement on the criteria of Annex IV referring to categories as in Article 9 of the AHL about WNF (Tables 13, 14, 15, 16 and 17). The expert judgement was based on ICBA approach described in detail in the opinion on the methodology. Experts have been provided with information of the disease fact-sheet mapped into Article 9 criteria (see supporting information, Annex A), based on that the experts indicate their Y/N or 'na' judgement on each criterion of Article 9, and the reasoning supporting their judgement.

The minimum number of judges in the judgement was 12. The expert judgement was conducted as described in the methodological opinion (EFSA AHAW Panel, 2017a). For details on the interpretation of the questions, see Appendix B of the methodological opinion (EFSA AHAW Panel, 2017a).

**Table 13:** Outcome of the expert judgement related to the criteria of Section 1 of Annex IV (category A of Article 9) for West Nile fever

| **Criteria to be met by the disease:** The disease needs to fulfil all of the following criteria | | **Final outcome** |
|---|---|---|
| 1 | The disease is not present in the territory of the Union OR present only in exceptional cases (irregular introductions) OR present in only in a very limited part of the territory of the Union | N |
| 2.1 | The disease is highly transmissible | N |
| 2.2 | There be possibilities of airborne or waterborne or vector-borne spread | Y |
| 2.3 | The disease affects multiple species of kept and wild animals OR single species of kept animals of economic importance | Y |
| 2.4 | The disease may result in high morbidity and significant mortality rates | N |
| **At least one criterion to be met by the disease:** In addition to the criteria set out above at points 1–2.4, the disease needs to fulfil at least one of the following criteria | | |
| 3 | The disease has a zoonotic potential with significant consequences on public health, including epidemic or pandemic potential OR possible significant threats to food safety | N |
| 4 (CI) | The disease has a significant impact on the economy of the Union, causing substantial costs, mainly related to its direct impact on the health and productivity of animals | N |
| 4 (PI) | The disease has a significant impact on the economy of the Union, causing substantial costs, mainly related to its direct impact on the health and productivity of animals | N |
| 5(a)(CI) | The disease has a significant impact on society, with in particular an impact on labour markets | N |
| 5(a)(PI) | The disease has a significant impact on society, with in particular an impact on labour markets | N |
| 5(b)(CI) | The disease has a significant impact on animal welfare, by causing suffering of large numbers of animals | N |
| 5(b)(PI) | The disease has a significant impact on animal welfare, by causing suffering of large numbers of animals | NC |
| 5(c)(CI) | The disease has a significant impact on the environment, due to the direct impact of the disease OR due to the measures taken to control it | N |
| 5(c)(PI) | The disease has a significant impact on the environment, due to the direct impact of the disease OR due to the measures taken to control it | NC |
| 5(d)(CI) | The disease has a significant impact on a long-term effect on biodiversity or the protection of endangered species or breeds, including the possible disappearance or long-term damage to those species or breeds | N |
| 5(d)(PI) | The disease has a significant impact on a long-term effect on biodiversity or the protection of endangered species or breeds, including the possible disappearance or long-term damage to those species or breeds | NC |

Colour code: green = consensus (Yes/No); yellow = no consensus (NC).

**Table 14:** Outcome of the expert judgement related to the criteria of Section 2 of Annex IV (category B of Article 9) for West Nile fever

| **Criteria to be met by the disease:** The disease needs to fulfil all of the following criteria | | **Final outcome** |
|---|---|---|
| 1 | The disease is present in the whole OR part of the Union territory with an endemic character AND (at the same time) several Member States or zones of the Union are free of the disease | Y |
| 2.1 | The disease is moderately to highly transmissible | Y |
| 2.2 | There be possibilities of airborne or waterborne or vector-borne spread | Y |
| 2.3 | The disease affects single or multiple species | Y |
| 2.4 | The disease may result in high morbidity with in general low mortality | Y |
| **At least one criterion to be met by the disease:** In addition to the criteria set out above at point 1–2.4, the disease needs to fulfil at least one of the following criteria | | |
| 3 | The disease has a zoonotic potential with significant consequences on public health, including epidemic potential OR possible significant threats to food safety | Y |
| 4 (CI) | The disease has a significant impact on the economy of the Union, causing substantial costs, mainly related to its direct impact on the health and productivity of animals | N |
| 4 (PI) | The disease has a significant impact on the economy of the Union, causing substantial costs, mainly related to its direct impact on the health and productivity of animals | N |
| 5(a)(CI) | The disease has a significant impact on society, with in particular an impact on labour markets | N |
| 5(a)(PI) | The disease has a significant impact on society, with in particular an impact on labour markets | N |
| 5(b)(CI) | The disease has a significant impact on animal welfare, by causing suffering of large numbers of animals | N |
| 5(b)(PI) | The disease has a significant impact on animal welfare, by causing suffering of large numbers of animals | NC |
| 5(c)(CI) | The disease has a significant impact on the environment, due to the direct impact of the disease OR due to the measures taken to control it | N |
| 5(c)(PI) | The disease has a significant impact on the environment, due to the direct impact of the disease OR due to the measures taken to control it | NC |
| 5(d)(CI) | The disease has a significant impact on a long-term effect on biodiversity or the protection of endangered species or breeds, including the possible disappearance or long-term damage to those species or breeds | N |
| 5(d)(PI) | The disease has a significant impact on a long-term effect on biodiversity or the protection of endangered species or breeds, including the possible disappearance or long-term damage to those species or breeds | NC |

Colour code: green = consensus (Yes/No); yellow = no consensus (NC).

**Table 15:** Outcome of the expert judgement related to the criteria of Section 3 of Annex IV (category C of Article 9) for West Nile fever

| **Criteria to be met by the disease:** The disease needs to fulfil all of the following criteria | | **Final outcome** |
|---|---|---|
| 1 | The disease is present in the whole OR part of the Union territory with an endemic character | Y |
| 2.1 | The disease is moderately to highly transmissible | Y |
| 2.2 | The disease is transmitted mainly by direct or indirect transmission | Y |
| 2.3 | The disease affects single or multiple species | Y |
| 2.4 | The disease usually does not result in high morbidity and has negligible or no mortality AND often the most observed effect of the disease is production loss | N |

| **At least one criterion to be met by the disease:** In addition to the criteria set out above at point 1–2.4, the disease needs to fulfil at least one of the following criteria | | |
|---|---|---|
| 3 | The disease has a zoonotic potential with significant consequences on public health, or possible significant threats to food safety | Y |
| 4(CI) | The disease has a significant impact on the economy of parts of the Union, mainly related to its direct impact on certain types of animal production systems | N |
| 4(PI) | The disease has a significant impact on the economy of parts of the Union, mainly related to its direct impact on certain types of animal production systems | N |
| 5(a)(CI) | The disease has a significant impact on society, with in particular an impact on labour markets | N |
| 5(a)(PI) | The disease has a significant impact on society, with in particular an impact on labour markets | N |
| 5(b)(CI) | The disease has a significant impact on animal welfare, by causing suffering of large numbers of animals | N |
| 5(b)(PI) | The disease has a significant impact on animal welfare, by causing suffering of large numbers of animals | NC |
| 5(c)(CI) | The disease has a significant impact on the environment, due to the direct impact of the disease OR due to the measures taken to control it | N |
| 5(c)(PI) | The disease has a significant impact on the environment, due to the direct impact of the disease OR due to the measures taken to control it | NC |
| 5(d)(CI) | The disease has a significant impact on a long-term effect on biodiversity or the protection of endangered species or breeds, including the possible disappearance or long-term damage to those species or breeds | N |
| 5(d)(PI) | The disease has a significant impact on a long-term effect on biodiversity or the protection of endangered species or breeds, including the possible disappearance or long-term damage to those species or breeds | NC |

Colour code: green = consensus (Yes/No); yellow = no consensus (NC).

**Table 16:** Outcome of the expert judgement related to the criteria of Section 4 of Annex IV (category D of Article 9) for West Nile fever

| **Criteria to be met by the disease:** The disease needs to fulfil all of the following criteria | | **Final outcome** |
|---|---|---|
| D | The risk posed by the disease in question can be effectively and proportionately mitigated by measures concerning movements of animals and products in order to prevent or limit its occurrence and spread | N |
| The disease fulfils criteria of Sections 1, 2, 3 or 5 of Annex IV of AHL | | Y |

Colour code: green = consensus (Yes/No).

**Table 17:** Outcome of the expert judgement related to the criteria of Section 5 of Annex IV (category E of Article 9) for West Nile fever

| Diseases in category **E need to fulfil criteria of Sections 1, 2 or 3 of Annex IV of AHL** and/or the following: | | **Final outcome** |
|---|---|---|
| E | Surveillance of the disease is necessary for reasons relating to animal health, animal welfare, human health, the economy, society or the environment (If a disease fulfils the criteria as in Article 5, thus being eligible to be listed, consequently category E would apply.) | Y |

Colour code: green = consensus (Yes/No).

### 3.3.1. Non-consensus questions

This section displays the assessment related to each criterion of Annex IV referring to the categories of Article 9 of the AHL where no consensus was achieved in form of tables (Tables 18, 19 and 20). The proportion of Y, N or 'na' answers are reported, followed by the list of different supporting views for each answer.

**Table 18:** Outcome of the expert judgement related to criterion 5(b)(PI) of Article 9

| Question | | Final outcome | Response | | |
|---|---|---|---|---|---|
| | | | Y (%) | N (%) | na (%) |
| 5(b) | The disease has a significant impact on animal welfare, by causing suffering of large numbers of animals | NC | 83 | 17 | 0 |

NC: non-consensus; number of judges: 12.

Reasoning supporting the judgement

Supporting Yes:

- WNF may have a potential impact on animal health and consequently welfare if introduced in naive populations in the absence of controls.
- The percentage of affected horses with severe clinical signs undergoes a significant impact on animal welfare.

Supporting No:

- Currently, there is no significant impact on welfare although the virus is circulating in many countries. It could have a potential impact in naïve populations.
- Although there has been extensive geographical expansion of the European territories with WNV circulation, the number of clinical cases in horses remains very limited.

**Table 19:** Outcome of the expert judgement related to criterion 5(c)(PI) of Article 9

| Question | | Final outcome | Response | | |
|---|---|---|---|---|---|
| | | | Y (%) | N (%) | na (%) |
| 5(c) | The disease has a significant impact on the environment, due to the direct impact of the disease OR due to the measures taken to control it | NC | 58 | 0 | 42 |

NC: non-consensus; number of judges: 12.

Reasoning supporting the judgement

Supporting Yes:

- The impact on wild birds could be potentially significant, if there was a substantial increase in outbreaks.
- Impacts of controls on the environment may be substantial.
- Supporting na:
- There are no data on the potential effect of vector control easures on the environment.

**Table 20:** Outcome of the expert judgement related to criterion 5(d)(PI) of Article 9

| Question | | Final outcome | Response | | |
|---|---|---|---|---|---|
| | | | Y (%) | N (%) | na (%) |
| 5(d) | The disease has a significant impact on a long-term effect on biodiversity or the protection of endangered species or breeds, including the possible disappearance or long-term damage to those species or breeds | NC | 67 | 33 | 0 |

NC: non-consensus; number of judges: 12.

Reasoning supporting the judgement

Supporting Yes:

- There may be a potential impact, given the range of bird species potentially affected. Some endangered species of, e.g. prey birds could disappear or be seriously threatened.
- The emergence of WNV strains more virulent for wild birds can affect larger numbers of animals.

Supporting No:

- There is an impact on individual animals, but no long-term effect on a population scale. Furthermore there are periodic epidemic outbreaks, but the probability of long-term epidemics in wild fauna is low.

### 3.3.2. Outcome of the assessment of criteria in Annex IV for West Nile fever for the purpose of categorisation as in Article 9 of the AHL

As from the legal text of the AHL, a disease is considered fitting in a certain category (A, B, C, D or E corresponding to point (a) to point (e) of Article 9(1) of the AHL) if it is eligible to be listed for Union intervention as laid down in Article 5(3) and fulfils all criteria of the first set from 1 to 2.4 and at least one of the second set of criteria from 3 to 5(d) as shown in Tables 11–17. According to the assessment methodology (EFSA AHAW Panel, 2017a), a criterion is considered fulfilled when the outcome is 'Yes'. With respect to different type of impact where the assessment is divided into current and potential impact, a criterion will be considered fulfilled if at least one of the two outcomes is 'Y' and, in case of no 'Y', the assessment is inconclusive if at least one outcome is 'NC'.

A description of the outcome of the assessment of criteria in Annex IV for WNF for the purpose of categorisation as in Article 9 of the AHL is presented in Table 21.

**Table 21:** Outcome of the assessment of criteria in Annex IV for WNF for the purpose of categorisation as in Article 9 of the AHL (CI = current impact; PI = potential impact)

| Category | Article 9 criteria | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | 1° set of criteria | | | | | 2° set of criteria | | | | | |
| | 1 | 2.1 | 2.2 | 2.3 | 2.4 | 3 | 4 | 5a | 5b | 5c | 5d |
| | Geographical distribution | Transmissibility | Routes of transmission | Multiple species | Morbidity and mortality | Zoonotic potential | Impact on economy | Impact on society | Impact on animal welfare | Impact on environment | Impact on biodiversity |
| A | N | N | Y | Y | N | N | N | N | CI: N PI: NC | CI: N PI: NC | CI: N PI: NC |
| B | Y | Y | Y | Y | Y | Y | N | N | CI: N PI: NC | CI: N PI: NC | CI: N PI: NC |
| C | Y | Y | Y | Y | N | Y | N | N | CI: N PI: NC | CI: N PI: NC | CI: N PI: NC |
| D | N | | | | | | | | | | |
| E | Y | | | | | | | | | | |

According to the assessment here performed, WNF complies with the following criteria of the Sections 1–5 of Annex IV of the AHL for the application of the disease prevention and control rules referred to in points (a)–(e) of Article 9(1):

1) To be assigned to category A, a disease needs to comply with all criteria of the first set (1, 2.1–2.4) and according to the assessment WNF complies with criteria 2.2 and 2.3, but not with 1, 2.1 and 2.4. To be eligible for category A, a disease needs to comply additionally with one of the criteria of the second set (3, 4, 5a–d) and WNF does not comply with criteria 3, 4 and 5a and the assessment is inconclusive on compliance with criteria 5b, 5c and 5d.
2) To be assigned to category B, a disease needs to comply with all criteria of the first set (1, 2.1–2.4) and according to the assessment WNF complies with all of them. To be eligible for category B, a disease needs to comply additionally with one of the criteria of the second

set (3, 4, 5a–d) and WNF complies with criterion 3, but not with criteria 4 and 5a and the assessment is inconclusive on compliance with criteria 5b, 5c and 5d.

3) To be assigned to category C, a disease needs to comply with all criteria of the first set (1, 2.1–2.4) and according to the assessment WNF complies with criteria 1, 2.1, 2.2 and 2.3, but not with 2.4. To be eligible for category C, a disease needs to comply additionally with one of the criteria of the second set (3, 4, 5a–d) and WNF complies with criterion 3, but not with criteria 4 and 5a and this assessment is inconclusive on compliance with criteria 5b, 5c and 5d.
4) To be assigned to category D, a disease needs to comply with criteria of Sections 1, 2, 3 or 5 of Annex IV of the AHL and with the specific criterion D of Section 4. WNF does not comply with the latter.
5) To be assigned to category E, a disease needs to comply with criteria of Sections 1, 2 or 3 of Annex IV of the AHL and/or the surveillance of the disease is necessary for reasons relating to animal health, animal welfare, human health, the economy, society or the environment. The latter is applicable if a disease fulfils the criteria as in Article 5, with which WNF complies.

### 3.4. Assessment of Article 8

This section presents the results of the assessment on the criteria of Article 8(3) of the AHL about WNF. The Article 8(3) criteria are about animal species to be listed, as it reads below:

'3. Animal species or groups of animal species shall be added to this list if they are affected or if they pose a risk for the spread of a specific listed disease because:

a) they are susceptible for a specific listed disease or scientific evidence indicates that such susceptibility is likely; or
b) they are vector species or reservoirs for that disease, or scientific evidence indicates that such role is likely'.

For this reason the assessment on Article 8 criteria is based on the evidence as extrapolated from the relevant criteria of Article 7, i.e. the ones related to susceptible and reservoir species or routes of transmission, which cover also possible role of biological or mechanical vectors.[2] According to the mapping, as presented in Table 5, Section 3.2 of the scientific opinion on the ad hoc methodology (EFSA AHAW Panel, 2017a), the main animal species to be listed for WNF according to the criteria of Article 8(3) are several species of birds and mammals, displayed in details in Table A.1 in Appendix A, as susceptible species. Several bird species belonging to the families of Corvidae, Passeridae and Fringillidae (order of Passeriformes) and to the family of Columbidae (order Columbiformes) can be considered reservoir species for WNV in Europe, details are shown in Table B.1 in Appendix B. The main vectors are some species of mosquitoes belonging to the genera *Culex*, *Aedes* and *Coquillettidia* (family Culicidae, order Diptera). The vector species are listed in https://efsa.maps.arcgis.com/apps/MapJournal/index.html?appid=512a03aa8df84d54a51bcb69d1b62735 (EFSA AHAW Panel, 2017b).

## 4. Conclusions

**TOR 1:** *for each of those diseases an assessment, following the criteria laid down in Article 7 of the AHL, on its eligibility of being listed for Union intervention as laid down in Article 5(3) of the AHL;*

- According to the assessment here performed, WNF complies with all criteria of the first set and with one criterion of the second set and therefore can be considered eligible to be listed for Union intervention as laid down in Article 5(3) of the AHL.

**TOR 2a:** *for each of the diseases which was found eligible to be listed for Union intervention, an assessment of its compliance with each of the criteria in Annex IV to the AHL for the purpose of categorisation of diseases in accordance with Article 9 of the AHL;*

- According to the assessment here performed, WNF meets the criteria as in Sections 2 and 5 of Annex IV of the AHL, for the application of the disease prevention and control rules referred to in points (b) and (e) of Article 9(1) of the AHL.

[2] A vector is a living organism that transmits an infectious agent from an infected animal to a human or another animal. Vectors are frequently arthropods. Biological vectors may carry pathogens that can multiply within their bodies and be delivered to new hosts, usually by biting. In mechanical vectors, the pathogens do not multiply within the vector, which usually remains infected for shorter time than in biological vectors.

**TOR 2b:** *for each of the diseases which was found eligible to be listed for Union intervention, a list of animal species that should be considered candidates for listing in accordance with Article 8 of the AHL.*

- According to the assessment here performed, the animal species that can be considered to be listed for WNF according to Article 8(3) of the AHL are, as susceptible species, several orders of birds and mammals and two orders of reptiles, as reported in Table A.1 in Appendix A of the present document. Reservoirs are several bird species belonging to the families of Corvidae, Passeridae and Fringillidae (order of Passeriformes) and to the family of Columbidae (order Columbiformes). Vectors are some species of mosquitoes belonging to the genera *Culex*, *Aedes* and *Coquillettidia* (family Culicidae, order Diptera).

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## Abbreviations

| | |
|---|---|
| ADNS | Animal Diseases Notification System |
| AHAW | EFSA Panel on Animal Health and Welfare |
| AHL | Animal Health Law |
| ALF | Alfuy |
| CDC | Centers for Disease Control and Prevention |
| CFSPH | Center for Food Security and Public Health |
| CITES | Convention on International Trade in Endangered Species of Wild Fauna and Flora |
| CPC | Cacipacore |
| DALY | disability-adjusted life year |

| | |
|---|---|
| ECDC | European Centre for Disease Prevention and Control |
| ELISA | enzyme-linked immunosorbent assay |
| HCDCP | Hellenic Center for Disease Control & Prevention |
| HI | haemagglutination inhibition |
| ICBA | Individual and Collective Behavioural Aggregation |
| IgG | immunoglobulin G |
| IgM | immunoglobulin M |
| ISS | Istituto Superiore di Sanità |
| IZSAM | Istituto Zooprofilattico Sperimentale dell'Abruzzo e del Molise |
| IUCN | International Union for Conservation of Nature |
| KOU | Koutango |
| KUN | Kunjin |
| LLIN | long lasting impregnated net |
| MS | Member State |
| MVE | Murray Valley encephalitis |
| OIE | World Organisation for Animal Health |
| PCR | polymerase chain reaction |
| p.i. | post infection |
| PFU | plaque-forming unit |
| PRN | plaque reduction neutralisation |
| RNA | ribonucleic acid |
| RT-PCR | reverse transcription polymerase chain reaction |
| SLE | St. Louis encephalitis |
| TBE | tick-borne encephalitis |
| $TCID_{50}$ | tissue culture infective dose, median |
| ToR | Terms of Reference |
| USU | Usutu |
| VN | virus neutralisation |
| WNF | West Nile fever |
| WNV | West Nile virus |
| YAO | Yaounde |

# Appendix A – Animal species infected naturally and experimentally by WNV

**Table A.1:** Naturally susceptible wildlife species (or family/orders)

| Class | Order | Family | Species |
|---|---|---|---|
| Aves | Anseriformes | Anatidae | Wood duck-*Aix sponsa*, Eurasian wigeon-*Anas penelope* (c), bronze-winged duck (spectacled duck)-*Anas specularis* (c), canvasback-*Aythya valisineria*, Canada goose-*Branta Canadensis*, barnacle goose-*Branta leucopsis* (c)(a), emperor goose-*Chen canagica* (c), greater Magellan goose (Andean goose)-*Chloephagapicta leucoptera* (c)(a), Abyssinian blue-winged goose-*Cyanochen cyanopterus* (c)(a), tundra swan-*Cygnus columbianus* (c), trumpeter swan-*Cygnus Cygnus buccinator* (c)(a), mute swan-*Cygnus olor*, Rosy-billed pichard-*Netta peposaca* (c)(a), ruddy duck-*Oxyura jamaicensis* |
| | Apodiformes | Apodidae | Chimney swift-*Chaetura pelagica* |
| | | Trochilidae | Ruby-throated hummingbird-*Archilochus colubris* |
| | Caprimulgiformes | Caprimulgidae | Common nighthawk-*Chordeiles minor* |
| | Casuariiformes | Dromaiidae | Emu-*Dromaius novaehollandiae* (c) |
| | Charadriiformes | Haradriidae | Ruddy turnstone-*Arenaria* interpres, killdeer-*Charadrius* vociferous, piping plover-*Charadrius melodus* |
| | | Laridae | European herring gull-*Larus argentatus*, laughing gull-*Larus atricilla*, ring-billed gull-*Larus delawarensis*, great black-backed gull-*Larus marinus,* black skimmer-*Rhynchops niger*, grey gull-*Larus modestus* (c)(a), Inca tern-*Larosterna inca* (c)(a) |
| | Ciconiformes | Ardeidae | Yellow-crowned night-heron-*Nyctanassa violacea* (c), black-crowned night-heron-*Nycticorax nycticorax* (c), great blue heron-*Ardea Herodias*, green heron-*Butorides virescens*, least bittern-*Ixobrychus exilis* |
| | | Cathartidae | Turkey vulture-*Cathartes aura*, black vulture-*Coragyps atratus*, king vulture-*Sarcoramphus papa* (c)(a) |
| | | Ciconiidae | Saddle-billed stork-*Ephippiorhynchos senegalensis* (c)(a), marabou stork-*Leptopilos crumeniferus* (c)(a), lesser adjutant-*Leptoptilos javanicus* (c)(a) |
| | | Phoenicopteridae | Chilean flamingo-*Phoenicopterus chilensis* (c), greater flamingo-*Phoenicopterus ruber ruber* (c) |
| | | Threskiornithidae | Scarlet ibis-*Eudocimus ruber* (c), northern bald ibis-*Geronticus eremita* (c)(a) |
| | Columbiformes | Columbidae | White-crowned pigeon-*Columba leucocephala*, rock dove-*Columba livia*, Mauritius pink pigeon-*Columba mayeri* (c)(a), common ground-dove-*Columbina passerine*, Eurasian collared-dove-*Streptopelia decaocto*, white-winged dove-*Zenaida asiatica*, mourning dove-*Zenaida macroura*, Luzon bleeding-heart-*Gallicolumba luzonica* (c)(a), Inca dove-*Columbina inca* |
| | Coraciiformes | Alcedinidae | Belted kingfisher-*Ceryle alcyon* |
| | Cuculiformes | Cuculidae | Yellow-billed cuckoo-*Coccyzus americanus* |

| Class | Order | Family | Species |
|---|---|---|---|
| | Falconiformes | Accipitridae | Cooper's hawk-*Accipiter cooperii*, Northern goshawk-*Accipiter gentilis*, sharp-shinned hawk-*Accipiter striatus*, golden eagle-*Aquila chrysaetos*, red-tailed hawk-*Buteo jamaicensis*, rough-legged hawk-*Buteo lagopus* (c), red-shouldered hawk-*Buteo lineatus*, broad-winged hawk-*Buteo platypterus*, Swainson's hawk-*Buteo swainsoni*, Northern harrier-*Circus cyaneus*, swallow-tailed kite-*Elanoides forficatus*, bald eagle-*Haliaeetus leucocephalus*, Mississippi kite-*Ictinia mississippiensis*, Osprey-*Pandion haliaetus*, Harris's hawk-*Parabuteo unicinctus* (c) |
| | | Falconidae | Merlin-*Falco columbarius*, prairie falcon-*Falco mexicanus*, peregrine falcon-*Falco peregrinus*, American kestrel-*Falco sparverius* |
| | Galliformes | Numididae | Crested guineafowl-*Guttera pucherani* (c)(a) |
| | | Odontophoridae | Northern bobwhite-*Colinus virginianus* |
| | | Phasianidae | Chukar-*Alectoris chukar* (c)(a), ruffed grouse-*Bonasa umbellus*, green junglefowl-*Gallus varius* (c)(a), Himalayan monal-*Lophophorus impeyanus* (c), Bulwer's pheasant-*Lophura bulweri* (c)(a), ring-necked pheasant-*Phasianus colchicus*, mountain peacock-pheasant-*Polypectron inopinatum* (c)(a), crested partridge-*Rollulus roulroul* (c)(a), Blyth's tragopan-*Tragopan blythii* (c), argus pheasant (unspecified)-various (c)(a), greater sage grouse-*Centrocerus urophasianus* |
| | Gaviiformes | Gaviidae | Common loon-*Gavia immer* |
| | Gruiformes | Gruidae | Demoiselle crane-*Anthropoides virgo* (c)(a), West African crowned crane-*Balearica pavonina pavonina* (a), wattled crane-*Bugeranus carunculatus* (c)(a), whooping crane-*Grus americana* (c)(a), Mississippi sandhill crane-*Grus canadensis pulla* (c), red-crowned crane-*Grus japonensis* (c)(a), Siberian crane-*Grus leucogeranus* (c)(a), hooded crane-*Grus monacha* (c)(a), white-naped crane-*Grus vipio* (c)(a) black-necked crane-*Grus nigricollis* (c)(a) |
| | | Rallidae | Virginia rail-*Rallus limicola* |
| | Musophagiformes | Musophagidae | Lady Ross's turaco-*Musophaga rossae* (c)(a) |
| | Passeriformes | Bombycillidae | Cedar waxwing-*Bombycilla cedrorum* |
| | | Cardinalidae | Northern cardinal-*Cardinalis cardinalis*, blue grosbeak-*Guiraca caerulea* (a), rose-breasted grosbeak-*Pheucticus ludovicianus*, dickcissel-*Spiza americana* |
| | | Corvidae | Western scrub-jay-*Aphelocoma californica*, American crow-*Corvus brachyrhynchos*, common raven-*Corvus corax*, fish crow-*Corvus ossifragus*, blue jay-*Cyanocitta cristata*, Steller's jay-*Cyanocitta stelleri*, black-billed magpie-*Pica hudsonia* (c) |
| | | Emberizidae | Song sparrow-*Melospiza melodia*, savannah sparrow-*Passerculus sandwichensis*, fox sparrow-*Passerella iliaca*, Eastern towhee-*Pipilo erythrophthalmus*, field sparrow-*Spizella pusilla* |
| | | Estrildidae | Zebra finch-*Taeniophygia guttata* (c) |

| Class | Order | Family | Species |
|---|---|---|---|
| | | Fringillidae | American goldfinch-*Carduelis tristis*, house finch-*Carpodacus mexicanus*, purple finch-*Carpodacus purpureus*, evening grosbeak-*Coccothraustes vespertinus*, European goldfinch-*Carduelis carduelis* (c) |
| | | Hirundinidae | Barn swallow-*Hirundo rustica*, purple martin-*Progne subis*, tree swallow-*Tachycineta bicolor* |
| | | Icteridae | Red-winged blackbird-*Agelaius phoeniceus*, rusty blackbird-*Euphagus carolinus*, Brewer's blackbird-*Euphagus cyanocephalus*, Baltimore oriole-*Icterus galbula*, brown-headed cowbird-*Molothrus ater*, boat-tailed grackle-*Quiscalus major*, great-tailed grackle-*Quiscalus mexicanus*, common grackle-*Quiscalus quiscula* |
| | | Laniidae | Loggerhead shrike-*Lanius ludovicianus* |
| | | Mimidae | Gray catbird-*Dumetella carolinensis*, Northern mockingbird-*Mimus polyglottos*, brown thrasher-*Toxostoma rufum* |
| | | Paridae | Tufted titmouse-*Baeolophus bicolor*, varied tit-*Parus varius* (c), black-capped chickadee-*Poecile atricapilla*, Carolina chickadee-*Poecile carolinensis* |
| | | Parulidae | Black-throated blue warbler-*Dendroica caerulescens*, yellow-rumped warbler-*Dendroica coronate*, yellow warbler-*Dendroica petechial*, blackpoll warbler-*Dendroica striata*, common yellowthroat-*Geothlypis trichas*, Kentucky warbler-*Oporornis formosus*, Northern parula-*Parula Americana*, ovenbird-*Seiurus aurocapillus*, Northern waterthrush-*Seiurus noveboracensis*, Nashville warbler-*Vermivora ruficapilla*, Canada warbler-*Wilsonia Canadensis*, hooded warbler-*Wilsonia citrina* |
| | | Passeridae | House sparrow-*Passer domesticus* |
| | | Sylviidae | White-crested laughingthrush-*Garrulax leucolophus* (c)(a) |
| | | Sittadae | White-breasted nuthatch-*Sitta carolinensis* |
| | | Sturnidae | European starling-*Sturnus vulgaris* |
| | | Thraupidae | Palm tanager-*Thraupis palmarum* (c) |
| | | Troglodytidae | Carolina wren-*Thryothaurus ludovicianus*, winter wren-*Troglodytes troglodytes* |
| | | Turdidae | Veery-*Catharus fuscescens*, hermit thrush-*Catharus guttatus*, gray-cheeked thrush-*Catharus minimus*, Swainson's thrush-*Catharus ustulatus*, wood thrush-*Hylocichla mustelina*, Eastern bluebird-*Sialia sialis*, American robin-*Turdus igratorius* |
| | | Tyrannidae | Traill's flycatcher-*Empidonax traillii/alnorum*, Eastern phoebe-*Sayornis phoebe*, scissor-tailed flycatcher-*Tyrannus forficatus*, Eastern kingbird-*Tyrannus tyrannus* |
| | | Vireonidae | Black-whiskered vireo-*Vireo altiloquus*, warbling vireo-*Vireo gilvus*, red-eyed vireo-*Vireo olivaceus* |
| | Pelecaniformes | Pelecanidae | American white pelican-*Pelecanus erythrorhynchos*, brown pelican-*Pelicanus occidentalis* (c)(a), double-crested cormorant-*Phalacrocorax auritus*, guanay cormorant-*Phalacrocorax bougainvillei* (c) |
| | Piciformes | Picidae | Red-headed woodpecker-*Melanerpes erythrocephalus*, downy woodpecker-*Picoides pubescens*, yellow-bellied sapsucker-*Sphyrapicus varius* |

| Class | Order | Family | Species |
|---|---|---|---|
| | Podicipediformes | Podicipedidae | Pied-billed grebe-*Podilymbus podiceps* |
| | Psittaciformes | Cacatuidae | Cockatoo (unspecified)-*Cacatua* spp. (c), cockatiel-*Nymphicus hollandicus* (c) |
| | | Psittacidae | Red-crowned parrot-*Amazona viridigenalis* (c), macaw (unspecified)-*Ara* spp. (c), budgerigar-*Melopsittacus undulatus* (c), lorikeet spp.-*Tricheglossus* spp. (c) |
| | Sphenisciformes | Spheniscidae | African penguin-*Spheniscus demersus* (c), Magellan penguin-*Spheniscus humboldti* (c)(a) |
| | Strigiformes | Strigidae | Northern saw-whet owl-*Aegolius acadicus*, boreal owl-*Aegolius funereous* (c), short-eared owl-*Asio flammeus*, Verreaux's eagle owl (milky eagle owl)-*Bubo lacteus* (c)(a), great horned owl-*Bubo virginianus*, snowy owl-*Nyctea scandiaca* (c), Eastern screech owl-*Otus asio*, tawny owl-*Strix aluco* (c), great grey owl-*Strix nebulosa* (c), spotted owl-*Strix occidentalis* (c), barred owl-*Strix varia,* Northern hawk owl-*Surnia ulula* (c) |
| | | Tytonidae | Barn owl-*Tyto alba* |
| | Struthioniformes | Struthionidae | Ostrich-*Struthio camelis* (c)(a) |
| Mammalia | Artiodactyla | Bovidae | Mountain goat-*Oreamnos americanus* (c) |
| | | Camelidae | Llama-*Lama glama* (c), alpaca-*Lama pacos* (c) |
| | | Cervidae | White-tailed deer-*Odocoileus virgninianus,* reindeer-*Rangifer tarnadus* (c), mule deer-*Odocoileus hemionus* |
| | | Suidae | Babirusa-*Babyrousa babyrousa* (c)(a) |
| | Carnivora | Canidae | Timber wolf-*Canis lupus* (c) |
| | | Mustelidae | Striped skunk-*Mephitis mephitis* |
| | | Phocidae | Harbor seal-*Phoca vitulina* (c) |
| | | Procyonidae | Red panda-*Ailurus fulgens fulgens* (c)(a) |
| | | Ursidae | Black bear-*Ursus americanus* (a) |
| | Chiroptera | Vespertilionidae | Big brown bat-*Eptesicus fuscus*, little brown bat-*Myotis lucifugus* |
| | Perissodactyla | Rhinocerotidae | Great Indian rhinoceros-*Rhinoceros unicornis* (c)(a) |
| | Primates | Cercopithecidae | Barbary macaque-*Macaca sylvanus* (c) |
| | | Lemuridae | Ring-tailed lemur-*Lemur catta* (c) |
| | Proboscidea | Elephantidae | Indian (Asian) elephant-*Elephas maximus indicus* (c)(a) |
| | Rodentia | Sciuridae | Gray squirrel-*Sciurus carolinensis*, fox squirrel-*Sciurus niger*, Eastern chipmunk-*Tamias striatus* |
| Reptilia | Crocodylia | Alligatoridae | American alligator-*Alligator mississippiensis* (c) |
| | Squamata | Varanidae | Crocodile monitor-*Varanus salvadorii* (c)(a) |

(c) Denotes either a captive or farmed animal(s). Virus or viral RNA was detected in animal tissue unless followed by an (a), which denotes detectable antibodies only have been reported (Source: USGS, National Wildlife Health Center (USGS, online)).

**Table A.2:** Naturally susceptible domestic species (or family/orders)

| Class | Order | Family | Species |
|---|---|---|---|
| Aves | Galliformes | Phasianidae | Domestic chicken (Red junglefowl)-*Gallus gallus*<br>Turkey (domestic and wild)-*Meleagris gallopavo* |
| | Anseriformes | Anatidae | Mallard-*Anas platyrhynchos*<br>Domestic goose-*Anser chinensis* (c)(a) |

| Class | Order | Family | Species |
|---|---|---|---|
| Mammalia | Artiodactyla | Bovidae | Domestic cattle-*Bos taurus*<br>Domestic (suffolk) sheep-*Ovis aries* |
| | Carnivora | Canidae | Domestic dog-*Canis familiaris* |
| | | Felidae | Domestic cat (feral)-*Felis catus* |
| | Lagomorpha | Leporidae | Domestic rabbit-*Oryctolagus cuniculus* |
| | Perissodactyla | Equidae | Domestic horse-*Equus equus przewalski caballus*<br>Donkey-*Equus asinus*<br>Mule |

(c) Denotes either a captive or farmed animal(s). Virus or viral RNA was detected in animal tissue unless followed by an (a), which denotes detectable antibodies only have been reported (Source: USGS, National Wildlife Health Center (USGS, online)).

**Table A.3:** Summary outcomes of experimental infections of West Nile virus performed in wild birds (adapted from Pérez-Ramírez et al. (2014))

| Order | Family | Species | Strain | Mortality | Viraemia | Distribution | References |
|---|---|---|---|---|---|---|---|
| Passeriformes | Turdidae | American robin (*Turdus migratorius*) | NY | < 20% | H | AM | Komar et al. (2003), VanDalen et al. (2013) |
| | | Swainson's thrush (*Catharus ustulatus*) | NY | < 20% | M | AM | Owen et al. (2006) |
| | | Clay-coloured thrush (*Turdus grayi*) | TEC/TAB | 20–50%/ < 20% | M | AM | Guerrero-Sánchez et al. (2011) |
| | Corvidae | Carrion crow (*Corvus corone*) | FR/ISR | 20–50%/ > 50% | L | EUR/ASIA | Dridi et al. (2013) |
| | | American crow (*Corvus brachyrhynchos*) | NY/TEX/MEX | > 50% | H | AM | McLean et al. (2001), Komar et al. (2003), Brault et al. (2004), Weingartl et al. (2004), Kinney et al. (2006), Kipp et al. (2006), Brault et al. (2007, 2011), Nemeth et al. (2011) |
| | | | KEN/KUN | 20–50%/ < 20% | M | | |
| | | Fish crow (*Corvus ossifragus*) | NY | > 50% | H | AM | Komar et al. (2003), Kipp et al. (2006), Nemeth et al. (2011) |
| | | Little raven (*Corvus mellori*) | NY | < 20% | M | OCE | Bingham et al. (2010) |
| | | | KUN | < 20% | L | | |
| | | Hooded crow (*Corvus cornix*) | EGY | > 50% | H | EUR/ASIA/ AFR | Work et al. (1955) |
| | | Western scrub-jay (*Aphelocoma californica*) | NY | > 50% | H | AM | Reisen et al. (2005) |
| | | Blue jay (*Cyanocitta cristata*) | NY | > 50% | H | AM | Komar et al. (2003), Weingartl et al. (2004) |

| Order | Family | Species | Strain | Mortality | Viraemia | Distribution | References |
|---|---|---|---|---|---|---|---|
| | | Black-billed magpie (*Pica hudsonia*) | NY | > 50% | H | AM | Komar et al. (2003) |
| | | Jungle crow (*Corvus macrorhynchos*) | NY | > 50% | H | ASIA | Shirafuji et al. (2008) |
| | Passeridae | House sparrow (*Passer domesticus*) | NY/CA/KEN/ EGY/TAB/ TEC/SP/IT09 | > 50% | H | WORLDWIDE | Work et al. (1955), Komar et al. (2003, 2005), Langevin et al. (2005), Reisen et al. (2005, 2006), Nemeth et al. (2008), LaPointe et al. (2009); Nemeth et al. (2009a,b), Brault et al. (2011), Guerrero-Sánchez et al. (2011), Wheeler et al. (2012), Del Amo et al. (2014) |
| | | | TEX/KUN/ IT08 | < 20% | M | | |
| | | | MEX | < 20% | L | | |
| | | Cape sparrow (*Passer melanurus*) | SA* | Und | L | AFR | McIntosh et al. (1969) |
| | Icteridae | Red-winged blackbird (*Agelaius phoeniceus*) | NY | < 20% | M/L | AM | Komar et al. (2003), Reisen and Hahn (2007), Nemeth et al.(2009b) |
| | | Brown-headed cowbird (*Molothrus ater*) | NY | < 20% | L | AM | Reisen et al. (2006), Reisen and Hahn (2007) |
| | | Brewer's blackbird (*Euphagus cyanocephalus*) | NY | < 20% | H | AM | Reisen et al. (2006), Reisen and Hahn (2007) |
| | | Tricolored blackbird (*Agelaius tricolor*) | NY | < 20% | H | AM | Reisen and Hahn (2007) |
| | | Common grackle (*Quiscalus quiscula*) | NY | 20–50% | H | AM | Komar et al. (2003) |
| | | Great-tailed grackle (*Quiscalus mexicanus*) | TAB/TEC | > 50%/ 20–50% | H | AM | Guerrero-Sánchez et al. (2011) |
| | | Bay-winged cowbird (*Agelaioides badius*) | ARG | < 20% | L | AM | Diaz et al. (2011) |
| | | Shiny cowbird (*Molothrus bonariensis*) | ARG | < 20% | L | AM | Diaz et al. (2011) |

| Order | Family | Species | Strain | Mortality | Viraemia | Distribution | References |
|---|---|---|---|---|---|---|---|
| | Emberizidae | Song sparrow (*Melospiza melodia*) | NY | < 20% | M | AM | Reisen and Fang (2007) |
| | | White-crowned sparrow (*Zonotrichia leucophrys*) | NY | Und | na | AM | Reisen et al. (2006) |
| | Fringillidae | Hawai'i 'amakihi (*Hemignathus virens*) | NY | 20–50% | H | AM | LaPointe et al. (2009) |
| | | House finch (*Haemorhous mexicanus*) | NY | > 50% | H | AM | Komar et al. (2003), Reisen et al. (2005), Fang and Reisen (2006), Reisen et al. (2006) |
| | Ploceidae | African masked weaver (*Ploceus velatus*) | SA* | Und | M | AFR | McIntosh et al. (1969) |
| | | Red-billed quelea (*Quelea quelea*) | SA* | Und | L | AFR | McIntosh et al. (1969) |
| | | Red bishop (*Euplectes orix*) | SA* | Und | M | AFR | McIntosh et al. (1969) |
| | Hirundinidae | Cliff swallow (*Petrochelidon pyrrhonota*) | NY | < 20% | M | AM | Oesterle et al. (2009, 2010) |
| | Mimidae | Gray catbird (*Dumetella carolinensis*) | NY | < 20% | M | AM | Owen et al. (2006) |
| | | Northern mockingbird (*Mimus polyglottos*) | NY | < 20% | H | AM | Komar et al. (2005) |
| | Sturnidae | European starling (*Sturnus vulgaris*) | NY | < 20% | M | WORLDWIDE | Komar et al. (2003); Reisen et al. (2006) |
| | Cardinalidae | Northern cardinal (*Cardinalis cardinalis*) | NY | < 20% | H | AM | Komar et al. (2005); Owen et al. (2012) |
| | Paridae | Tufted titmouse (*Baeolophus bicolor*) | NY | > 50% | H | AM | Kilpatrick et al. (2013) |
| | Troglodytidae | Carolina wren (*Thryothorus ludovicianus*) | NY | 20–50% | H | AM | Kilpatrick et al. (2013) |
| Falconiformes | Falconidae | Gyrfalcon (*Falco rusticolus*) | AUS* | 20–50% | H | AM/EUR/AS | Ziegler et al. (2013) |
| | | | NY | 20–50% | M | | |
| | | Hybrid falcon (*Falco rusticolus x Falco cherrug*) | NY | < 20% | L | WORLDWIDE | Busquets et al. (2012) |
| | | American kestrel (*Falco sparverius*) | NY | < 20% | H | AM | Komar et al. (2003), Nemeth et al. (2006a) |
| | | Common kestrel (*Falco tinnunculus*) | EGY | < 20% | L | EUR/AS/AFR | Work et al. (1955) |

| Order | Family | Species | Strain | Mortality | Viraemia | Distribution | References |
|---|---|---|---|---|---|---|---|
| Accipitriformes | Accipitridae | Red-tailed hawk (*Buteo jamaicensis*) | NY | < 20% | H | AM | Nemeth et al. (2006a) |
| Strigiformes | Tytonidae | Barn owl (*Tyto alba*) | NY | < 20% | L | WORLDWIDE | Nemeth et al. (2006a) |
| | Strigidae | Great horned owl (*Bubo virginianus*) | NY | < 20% | H | AM | Komar et al. (2003), Nemeth et al. (2006a) |
| | | Eastern screech-owl (*Megascops asio*) | NY | > 50% | H | AM | Nemeth et al. (2006a) |
| Galliformes | Odontophoridae | California quail (*Callipepla californica*) | NY | < 20% | L | AM | Reisen et al. (2005, 2006) |
| | | Gambel's quail (*Callipepla gambelii*) | NY | < 20% | L | AM | Reisen et al. (2006) |
| | | Northern bobwhite (*Colinus virginianus*) | NY | < 20% | L | AM | Komar et al. (2003) |
| | Phasianidae | Red-legged partridge (*Alectoris rufa*) | SP/MO | 20–50%/ > 50% | H | EUR | Sotelo et al. (2011b) |
| | | | NY | > 50% | L | | Escribano-Romero et al. (2013) |
| | | Japanese quail (*Coturnix japonica*) | NY | < 20% | L | WORLDWIDE | Komar et al. (2003) |
| | | Ring-necked pheasant (*Phasianus colchicus*) | NY | < 20% | L | WORLDWIDE | Komar et al.(2003) |
| | | Greater sage-grouse (*Centrocercus urophasianus*) | NY | > 50% | M | AM | Clark et al. (2006) |
| Pelecaniformes | Ardeidae | Rufous night-heron (*Nycticorax caledonicus*) | KUN | < 20% | L | OCE | Boyle et al. (1983b,a) |
| | | Little egret (*Egretta garzetta*) | KUN | < 20% | L | EUR/AS/AFR/OCE | Boyle et al. (1983a,b) |
| | | Intermediate heron (*Mesophoyx intermedia*) | KUN | < 20% | L | AFR/AS | Boyle et al. (1983a,b) |
| | | Cattle egret (*Bubulcus ibis*) | SA*/EGY | Und/ < 20% | L | WORLDWIDE | Work et al. (1955); McIntosh et al. (1969) |
| | Threskiornithidae | African sacred ibis (*Threskiornis aethiopicus*) | SA* | Und | L | AFR/AS | McIntosh et al. (1969) |
| Columbiformes | Columbidae | Rock pigeon (*Columba livia*) | SA*/NY/TEC/TAB | Und/ < 20% | L | WORLDWIDE | McIntosh et al. (1969); Guerrero-Sánchez et al. (2011) |

| Order | Family | Species | Strain | Mortality | Viraemia | Distribution | References |
|---|---|---|---|---|---|---|---|
| | | Ring-necked dove (*Streptopelia capicola*) | SA* | Und | L | AFR | McIntosh et al. (1969) |
| | | Eurasian collared-dove (*Streptopelia decaocto*) | NY/CO | < 20%/ < 20% | M | AM/EUR/AS/ AFR | Panella et al. (2013) |
| | | Laughing dove (*Spilopelia senegalensis*) | SA*/EGY | Und/ < 20% | L | AFR/AS | Work et al. (1955), McIntosh et al. (1969) |
| | | Common ground-dove (*Columbina passerina*) | NY | Und | na | AM | Reisen et al. (2006, 2008) |
| | | Mourning dove (*Zenaida macroura*) | NY | < 20% | M | AM | Komar et al. (2003), Reisen et al. (2005, 2006) |
| | | Picui ground-dove (*Columbina picui*) | ARG | < 20% | M | AM | Diaz et al. (2011) |
| Gruiformes | Rallidae | American coot (*Fulica americana*) | NY | < 20% | L | AM | Komar et al. (2003) |
| | | Crested coot (*Fulica cristata*) | SA* | Und | L | AFR/EUR | McIntosh et al. (1969) |
| | Gruidae | Sandhill crane (*Grus canadensis*) | NY | < 20% | L | AM | Olsen et al. (2009) |
| Anseriformes | Anatidae | Common goose (*Anser anser*) | SA* | > 50% | M | WORLDWIDE | Banet-Noach et al. (2003) |
| | | Canada goose (*Branta canadensis*) | NY | < 20% | M | AM/EUR | Komar et al. (2003) |
| | | Mallard (*Anas platyrhynchos*) | NY | < 20% | H | WORLDWIDE | Komar et al. (2003) |
| | | Yellow-billed duck (*Anas undulata*) | SA* | Und | L | AFR | McIntosh et al. (1969) |
| | | Red-billed teal (*Anas erythrorhyncha*) | SA* | Und | L | AFR | McIntosh et al. (1969) |
| | | Southern pochard (*Netta erythrophthalma*) | SA* | Und | L | AFR | McIntosh et al. (1969) |
| Charadriiformes | Charadriidae | Killdeer (*Charadrius vociferus*) | NY | < 20% | H | AM | Komar et al. (2003) |
| | Laridae | Ring-billed gull (*Larus delawarensis*) | NY | > 50% | H | AM | Komar et al. (2003) |
| Psittaciformes | Psittacidae | Monk parakeet (*Myiopsitta monachus*) | NY | < 20% | L | AM | Komar et al. (2003) |
| | | Budgerigar (*Melopsittacus undulatus*) | NY | < 20% | L | OCE | Komar et al. (2003) |

| Order | Family | Species | Strain | Mortality | Viraemia | Distribution | References |
|---|---|---|---|---|---|---|---|
| Piciformes | Picidae | Northern flicker (*Colaptes auratus*) | NY | < 20% | M | AM | Komar et al. (2003) |

CA: California 04; NY: New York 99; CO: Colorado 08; SA: South Africa; ARG: Argentina 06; EGY: Egypt; KUN: Kunjin; SP: Spain 07; MO: Morocco 03; AUS: Austria 09; MEX: Mexico 03; TEX: Texas 03; KEN: Kenya 3829; FR: France 00; ISR: Israel 98; TEC: Tecato (Mexico); TAB: Tabasco (Mexico); IT08: Italy 08; IT09: Italy 09.* Lineage 2.
**L**: Low viraemia (mean peak viraemia ≤ 104 PFU/mL); **M**: Medium viraemia (mean peak viraemia 104–106 PFU/mL); **H**: High viraemia (mean peak viraemia > 106 PFU/mL); **na**: Data not available.
AFR: Africa; AM: America; AS: Asia; EUR: Europe; OCE: Oceania.
Und: Undetermined.

**Table A.4:** Summary outcomes of systematic review of experimental infections of domestic animals with WNV (papers published up to January 2016)

| Species | References | Number of animal groups[(a)] | Agent detection[(b)] | | Observation of clinical signs[(c)] | | Clinical signs (and number of groups in which were reported) |
|---|---|---|---|---|---|---|---|
| | | | Min day | Max day | Min day | Max day | |
| Cats | Austgen et al. (2004) | 3 (19 animals) | Virus isolation from blood: 1 (0.5–3) | Virus isolation from blood: 7 (4.5–8) | 1 | 6 | No clinical signs observed (2), fever (1), depression/ apathy (1) |
| | | | | | | | 0 dead animals |
| Dogs | Austgen et al. (2004), Karaca et al. (2005) | 2 (19 animals) | Virus isolation from blood: 1.3 (0.5–2) | Virus isolation from blood: 5.3 (4.5–6) | 1 | 1 | No clinical signs observed (1), fever (1) |
| | | | | | | | 0 dead animals |
| Horses | Bunning et al. (2002), Shirafuji et al. (2009), Castillo-Olivares et al. (2011) | 4 (17 animals) | Virus isolation from blood (3 groups): 3(1–4) | Virus isolation from blood (3 groups): 6 (6–7) | 6.5 (3–8) | 10 (9–11) | No clinical signs observed (1), twitching/tremors (1), neurological signs (2), fever (1) |
| | | | PCR from blood (1 group): 3 | PCR from blood (1 group): 7 | | | 1 dead animal in 1 group |
| Pigs | Teehee et al. (2005) | 2 (12 animals) | Virus isolation from blood: 1.5 (1.5–4.5) | Virus isolation from blood: 5 (4.5–5) | Not reported | | No clinical signs observed (1), not reported (1) |
| | | | | | | | 0 dead animals |
| Rabbits | Suen et al. (2015) | 2 (27 animals) | Not reported | | 1 | Not reported | No clinical signs observed (1), fever (1) |
| | | | | | | | 0 dead animals |
| Sheep | Barnard and Voges (1986) | 1 (2 animals) | Virus isolation from blood: 3 | Virus isolation from blood: 11 | 3 | 3 | Fever |

(a): ll data were analysed at animal group level, reflecting the animal groups followed and reported in the individual references. Some references reported more than one animal group.
(b): Min = first day (in dpi) that pathogen/RNA was detected in a sample for each reported animal group; Max = last day (in dpi) that virus/RNA was detected in a sample for each reported animal group. Min and Max were recorded individually for each animal group, and median (min-max) for each of those values were calculated from all group data (each group representing one observation, with no weighting based on the size of the animal groups). Contact transmission groups were not included in the summary.
(c): Min = first day (in dpi) in which clinical signs were observed in each whole animal group reported; Max = last day (in dpi) in which clinical signs were observed in each whole animal group reported. Min and Max were recorded individually for each animal group, and median (min-max) for each of those values were calculated from all group data (each group representing one observation, with no weighting based on the size of the animal groups). Contact transmission groups were not included in the summary.

# Appendix B – List of wild and domestic WNV reservoir/sentinel animal species

**Table B.1:** List of wild and domestic WNV reservoir/sentinel animal species

| Family | Reservoir | Sentinel | Notes |
|---|---|---|---|
| Turdidae | ND | Y | Intense viraemia and clinical signs developed by infected birds |
| Corvidae | Potential | Y | Intense viraemia and clinical signs developed by the infected birds with high mortality |
| Passeridae | Y | Y | Intense and long viraemia and clinical signs developed by infected birds |
| Anatidae | – | Y | Intense viraemia and clinical signs developed by infected birds |
| Columbidae | Y | – | Common ground-dove (*Columbina passerina*): WNV detection in spleen and kidney and lung at > 6 weeks p.i |
| Fringillidae | Y | – | Persistent infection in house finches (*Haemorhous mexicanus*) |
| Falconidae | – | Y | Intense viraemia and clinical signs developed by infected birds |
| Phasianidae | – | Y | Viraemia short and scarce, asymptomatic infection, detectable serological response |
| Laridae | – | Y | Intense viraemia and clinical signs developed by infected birds |
| Strigidae | | Y | Intense viraemia and clinical signs developed by infected birds |
| Equidae | – | Y | Viraemia short and scarce, development of clinical symptoms, detectable serological response |
| Canidae | – | Potential | Viraemia short and scarce, rare development of clinical symptoms, detectable serological response. Potential use as sentinel in urban areas |
| Felidae | – | Potential | Viraemia short and scarce, rare development of clinical symptoms, detectable serological response. Potential use as sentinel in urban areas |

# Appendix C – WNV morbidity and mortality rates in horses

**Table C.1:** WNV morbidity and mortality rates in horses (2010–2016 EU outbreaks)

| Outbreaks in equids | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| **Country** | **Year** | **No outbreaks** | **No outbreaks with clinical symptoms** | **No horses present** | **No total cases** | **No horses with symptoms** | **Died/ culled** | **Prevalence of infection** | **Case-morbidity rate** | **Case-fatality rate** |
| Italy | 2008 | 273 | 18 | 1,941 | 563 | 32 | 5 | 29% | 2% | 1% |
| | 2009 | 137 | 32 | 1,398 | 223 | 37 | 9 | 16% | 3% | 24% |
| | 2010 | 67 | 11 | 415 | 128 | 11 | 5 | 31% | 3% | 45% |
| | 2011 | 91 | 41 | 881 | 197 | 58 | 14 | 22% | 7% | 24% |
| | 2012 | 30 | 13 | 313 | 63 | 15 | 3 | 20% | 24% | 20% |
| | 2013 | 35 | 11 | 308 | 50 | 12 | 1 | 16% | 24% | 8% |
| | 2014 | 17 | 6 | 257 | 27 | 6 | 2 | 11% | 22% | 33% |
| | 2015 | 26 | 6 | 302 | 30 | 6 | 5 | 10% | 20% | 17% |
| | 2016* | 33 | 13 | 310 | 37 | 13 | 4 | 7% | 35% | 11% |
| Portugal | 2016 | 1 | 1 | 2 | 1 | 1 | 0 | 50% | 50% | 0% |
| | 2015 | 3 | 3 | 82 | 4 | 4 | 0 | 5% | 5% | 0% |
| | 2010 | 2 | 2 | 71 | 2 | 2 | 1 | 3% | 3% | 1% |
| Spain | 2011 | 5 | Unknown | 44 | 11 | Unknown | 1 | 25% | Unknown | 9% |
| | 2010 | 31 | 2 | 845 | 39 | 2 | 2 | 4% | 0% | 5% |
| France | 2015 | 35 | 26 | 262 | 49 | 34 | 5 | 19% | 13% | 0–5, 26% |
| | 2006 | 4 | 1 | 63 | 4 | 1 | 1 | 6% | 2% | 25% |
| Croatia | 2014 | 1 | 0 | 2 | 1 | 0 | 0 | 50% | 0% | 0% |
| | 2012 | 11 | 0 | 87 | 12 | 0 | 0 | 14% | 0% | 0% |
| Greece | 2014 | 4 | 0 | 51 | 4 | 0 | 0 | 8% | 0% | 0% |
| | 2013 | 10 | 2 | 559 | 15 | 2 | 1 | 3% | 0% | 7% |
| | 2012 | 14 | 3 | 100 | 15 | 3 | 0 | 15% | 3% | 0% |
| | 2011 | 17 | 0 | 374 | 23 | 0 | 1** | 6% | 0% | 0% |
| | 2010 | 27 | 3 | 559 | 30 | 3 | 3 | 5% | 1% | 10% |
| Romania | 2010 | 3 | Unknown | 9 | 6 | Unknown | 0 | 67% | Unknown | Unknown |
| Former Yugoslav Republic of Macedonia | 2011 | 4 | 0 | 51 | 10 | 0 | 0 | 20% | 0% | 0% |
| Bulgaria | 2010 | 2 | 0 | 118 | 8 | 0 | 0 | 7% | 0% | 0% |

*: 2016 Italian data: updated to 14 October 2016.
**: Death may have been the result of conditions other than West Nile virus infection (possible snake bite reported).
Source: Italian National information system and (OIE, online).